*Journal of Experimental Botany*, Vol. 69, No. 13 pp. 3267–3277, 2018
doi:10.1093/jxb/ery082 Advance Access publication 7 March 2018

REVIEW PAPER

# Genetic diversity of root system architecture in response to drought stress in grain legumes

**Heng Ye[1,*], Manish Roorkiwal[2,*], Babu Valliyodan[1,*], Lijuan Zhou[1], Pengyin Chen[3], Rajeev K. Varshney[2] and Henry T. Nguyen[1,†]**

[1] Division of Plant Sciences, University of Missouri, Columbia, MO 65211, USA
[2] Center of Excellence in Genomics & Systems Biology (CEGSB), International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patancheru, Telangana-502324, India
[3] Division of Plant Sciences, University of Missouri-Fisher Delta Research Center, Portageville, MO 63873, USA

* These authors contributed equally to this work.
† Correspondence: nguyenhenry@missouri.edu

Received 13 October 2017; Editorial decision 21 February 2018; Accepted 5 March 2018

Editor: Jianhua Zhang, Hong Kong Baptist University, Hong Kong

## Abstract

**Climate change has increased the occurrence of extreme weather patterns globally, causing significant reductions in crop production, and hence threatening food security. In order to meet the food demand of the growing world population, a faster rate of genetic gains leading to productivity enhancement for major crops is required. Grain legumes are an essential commodity in optimal human diets and animal feed because of their unique nutritional composition. Currently, limited water is a major constraint in grain legume production. Root system architecture (RSA) is an important developmental and agronomic trait, which plays vital roles in plant adaptation and productivity under water-limited environments. A deep and proliferative root system helps extract sufficient water and nutrients under these stress conditions. The integrated genetics and genomics approach to dissect molecular processes from genome to phenome is key to achieve increased water capture and use efficiency through developing better root systems. Success in crop improvement under drought depends on discovery and utilization of genetic variations existing in the germplasm. In this review, we summarize current progress in the genetic diversity in major legume crops, quantitative trait loci (QTLs) associated with RSA, and the importance and applications of recent discoveries associated with the beneficial root traits towards better RSA for enhanced drought tolerance and yield.**

**Keywords:** Climate change, drought tolerance, genomics, genomics-assisted breeding, legumes, natural variation.

## Introduction

Climate change has increased the frequency of extreme weather patterns including irregular precipitation, which can cause drought stress and adversely affect crop production (Lesk *et al.*, 2016). Yield losses of major crops under such fluctuating weather patterns are increasing, despite the progressive efforts in yield improvement through breeding and management practices since the 1960s (Boyer, 1983; Lobell *et al.*, 2014; Rosenzweig *et al.*, 2014). Estimates show that crop yields would decline 30–82% by the end of the 21st century under the current climate change, despite increasing concentrations of $CO_2$ that could benefit photosynthesis (Hatfield *et al.*, 2011).

Legumes form a major part of the human diet and animal feed, and are used for biofuel due to their oil content. Besides food and nutritional security, legumes act as a major cash crop, generating US$31 billion per annum to a large number of smallholders (>700 million) in developing nations (Abate et al., 2012). Legumes are known not only as an affordable protein-rich source, but also for their environmental and economic value. Inclusion of legumes in crop rotation programs leads to a positive environmental effect by improving the soil nutritional profile via symbiotic nitrogen fixation; it aids in sustainable agricultural production by promoting the yield of subsequent crops as well. It has been reported that when grown in rotation with cereals, in addition to improving the soil nutritional profile, they prevent soil erosion, and further lead to reduced incidences of soil pathogens (Daryanto et al., 2015). However, only a limited yield increase of 0–2% per year is occurring in legume crops (Foyer et al., 2016). Abruptly changing climatic conditions are having a negative impact on legume production. For example, soybean yields in the USA declined by 2–4% for every degree rise in temperature during the growing season, resulting a loss of US$11 billion (Mourtzinis et al., 2015). Rising global temperatures were reported to reduce the areas suitable for bean production due to increased drought stress (Beebe et al., 2011). Yields will decrease in developing countries in the tropics and subtropics such as India, Pakistan, and Ethiopia due to the increased occurrence of drought stress (Andrews and Hodge, 2010). A large yield gap for legume crops exists in Africa of >300% due to water-limiting issues. Moreover, as an integral part of Indian agriculture, legume crop production has remained unstable, with a yield gap for soybean ranging from 850–1320 kg $ha^{-1}$, for groundnut from 1180–2010 kg $ha^{-1}$, for pigeon pea from 550–770 kg $ha^{-1}$, and for chickpea from 610–1150 kg $ha^{-1}$ (Bhatia et al., 2006; Anderson et al., 2016).

Plants have evolved stress adaptation mechanisms under natural selection. Exotic and wild relatives tend to have better stress-resistant characteristics and maintain natural variations favorable for stress resistance (Mickelbart et al., 2015). Incorporation of these favorable natural variations into current elite germplasm is essential to improve the performance of cultivars under stress conditions. This can be achieved through the identification of resistant genetic resources, discovery of favorable natural variations, and finally the incorporation of these natural variations into the elite cultivars. These natural variations can be identified through quantitative trait locus (QTL) mapping and genome-wide association studies (GWAS) (Takeda and Matsuoka, 2008; Varshney et al., 2015). With the assistance of genomic strategies, these natural genetic variations can be identified and expeditiously incorporated into the cultivars by marker-assisted selection (MAS) and genomic selection strategies. Characterization of the candidate genes underlying these natural variations will further elucidate biological mechanisms, which consequently will be translated to crop improvement through genetic modifications.

Drought tolerance refers to the ability of plants to survive, grow, and reproduce under water deficit conditions (Levitt, 1972; Turner et al., 1978). In crops, drought tolerance actually suggests the ability of plants to yield under stress conditions (Turner, 2000; Fleury et al., 2010). Crop yield was determined by three major components: water use, water use efficiency, and harvest index (yield=water use×water use efficiency×harvest index) (Passioura, 1977, 1983; Turner, 2000; Turner et al., 2001). Studies clearly show a positive correlation between the amount of water reduction and yield reduction, whereas the degree of this correlation varies within legume species and the developmental stages at which plant encountered the drought stress (Daryanto et al., 2015). Acquisition of more water resource in plants is a key solution to improve drought tolerance in crops (Passioura, 1977, 1983). Thus, the development and distribution of the root system can be regarded as key factors for more efficient water uptake and thereby for managing the performance of legumes under drought stress (Newman and Moser, 1988; Gaur et al., 2008). Root traits are usually described as root system architecture (RSA), referring to the shape and physical space of the roots. A deeper and proliferative root system is able to avoid drought stress by its ability to acquire more water resource. RSA is also highly plastic to environmental changes, including water deficit stress. When plants perceive water deficit stress, roots tend to keep growing and penetrate into deeper soil layers (Hoogenboom et al., 1987; Sponchiado et al., 1989; Creelman et al., 1990; Wu et al., 1994).

## Root system architecture in drought tolerance

Fifty decades ago, Kramer (1969) stated one of the essential characteristics of drought tolerance: 'deep, wide-spreading, much-branched root system'. In order to access enhanced available soil moisture, plant adapts to greater rooting depth and root biomass (Blum, 2011; Fenta et al., 2014; Fig. 1). The benefit of a deep and proliferative root system for drought tolerance has been reported in various crops, including rice (Nguyen et al., 1997; Babu et al., 2003; Kato et al., 2006; MacMillan et al., 2006; Steele et al., 2006; Bernier et al., 2009; Uga et al., 2013), maize (Tuberosa et al., 2002; Giuliani et al., 2005; Hammer et al., 2009; Landi et al., 2010; Hund et al., 2011), barley (Forster et al., 2005), wheat (Manschadi et al., 2006; Wasson et al., 2012), chickpea (Varshney et al., 2011; Chen et al., 2012), and soybean (Hudak and Patterson, 1995; Pantalone et al., 1996; Sadok and Sinclair, 2011). The diameter and distribution of the metaxylem vessels that govern root conductivity are also known to provide drought tolerance in legumes (Purushothaman et al., 2013).

Deep rooting is a complex trait affected by growth angle and root length (Araki et al., 2002; Fig. 1). Root angle determines the direction of horizontal and vertical distribution of roots in the soil, and is recognized as an essential trait for drought avoidance in sorghum (Mace et al., 2012), wheat (Christopher et al., 2013), and rice (Uga et al., 2013). Root angle was correlated with rooting depth in rice (Kato et al., 2006), chickpea (Sayar et al., 2007; Kashiwagi et al., 2015), and sorghum (Singh et al., 2011). Wider root angles could reduce the energy inputs to penetrate in deeper horizons to access water during limited rainfall (Wasson et al., 2012; Meister et al., 2014). A natural genetic variation in root angle was cloned in rice as *DEEPER ROOTING 1* (*DRO1*) (Uga et al., 2013). *Dro1* doubled the yield

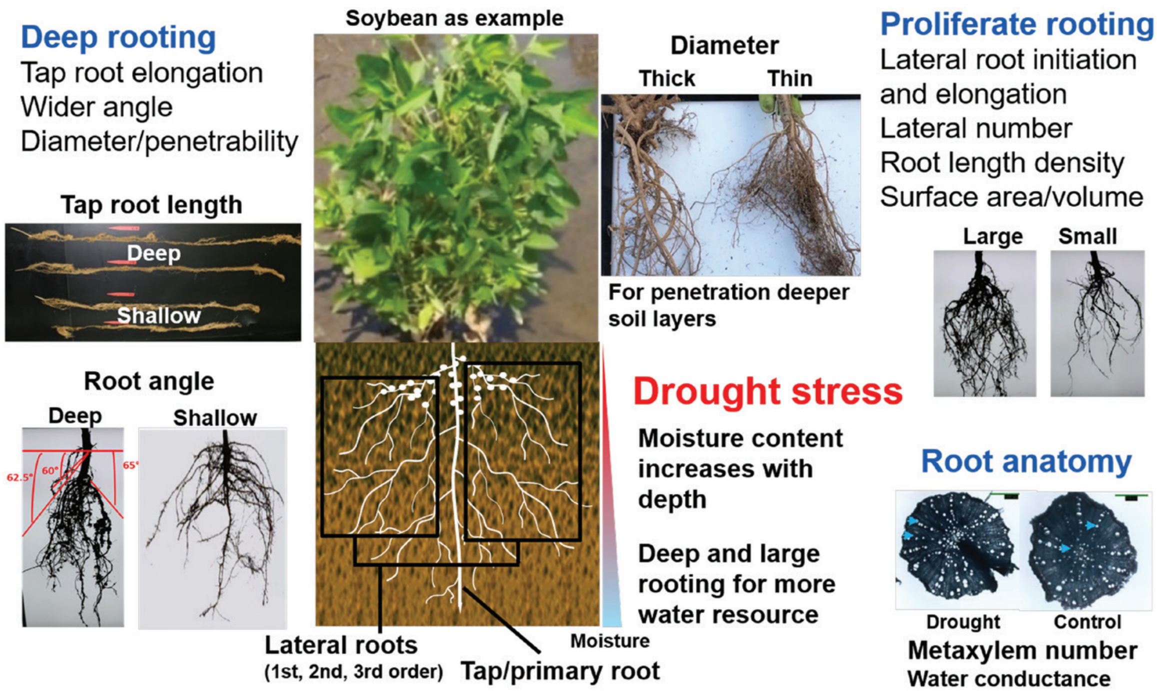


**Fig. 1.** Roles of root system architecture in the improvement of drought tolerance.

under moderate and severe drought stress due to its ability to develop a deep rooting system (Uga *et al.*, 2013). Another deep rooting determinant is root diameter/thickness and penetrability (Fig. 1). Thicker roots tend to penetrate deeper through soil layers for deep rooting (Yu *et al.*, 1995; Zheng *et al.*, 2000). Proliferative rooting is mainly determined by lateral root initiation and elongation, which usually refers to lateral root number, root length density, and root surface area and volume (Fig. 1). Proliferative roots have relatively high water uptake efficiency in water deficit soils. Chickpea lines with higher root length density have shown clearly a better performance in yield and drought tolerance-related traits under water deficit environments (Jaganathan *et al.*, 2015).

In addition to deep and proliferative rooting, drought stress also induces the plasticity responses of root systems by increasing the number of fibrous roots, and decreasing lateral root diameter and fluctuations in root biomass (Nielsen *et al.*, 1997; Osmont *et al.*, 2007; Meister *et al.*, 2014; Salazar-Henao *et al.*, 2016). Drought-tolerant lines tend to elongate their rooting depth significantly more than sensitive lines under drought stress in legumes (Garay and Wilhelm, 1983; Sponchiado *et al.*, 1989). Alterations in root anatomy, such as aerenchyma formation in maize (Lynch, 2011; Burton *et al.*, 2013) and soybean (Prince *et al.*, 2017), saves the energy inputs to allow improved soil penetration and exploration to mitigate water deficit (Addington *et al.*, 2006; Maseda and Fernández, 2006). The number of metaxylems was identified to be correlated with drought tolerance in soybean, and more metaxylems help in water transportation in the roots (Prince *et al.*, 2017; Fig. 1).

## Root phenotyping and modeling RSA

Root phenotyping in the laboratory, greenhouse, and field encompasses simple agar plates and germination papers, labor-intensive root digging in the field, an underground root observation facility, and sophisticated computer-assisted root imaging equipment (reviewed by Paez-Garcia *et al.*, 2015). Primary RSA traits include primary root length, root length density, root angle, fibrous and lateral roots, root diameters, and root anatomy. In laboratory conditions, a larger numbers of samples under well-controlled conditions can be phenotyped; however, RSA may be affected by the containers and cannot reflect real growth stages and environmental conditions. Greenhouse evaluation is an intermediate system between the laboratory and field. However, it has space limitations and requires more labor input. Field evaluation reflects the real performance of roots, correlating plant physiology with RSA well; however, intensive investment in labor is needed. Currently, high-throughput phenotyping of RSA has focused on field-based approaches in crops such as maize (York and Lynch, 2015), rice (Wissuwa *et al.*, 2016), and wheat (Richard *et al.*, 2015). With a recent shift from studying vegetative stage root traits in the greenhouse (Manavalan *et al.*, 2015; Prince *et al.*, 2015) to more field-based reproductive stage research (Burridge *et al.*, 2016) in legume species, knowledge of the genetic regulation of RSA has improved. The challenges of digging for a large number of plants using a 'Shovelomics' (Trachsel *et al.*, 2011) approach have been offset by the development and application of high-throughput image analysis platforms such as digital imaging of root traits (DIRT) (Bucksch *et al.*, 2014; Das *et al.*, 2015). The adoption of digital image analysis platforms for field-grown plants is gaining prevalence in legumes.

Mechanistic understanding of RSA and plasticity can be developed from functional–structural plant modeling systems. These modeling systems rely on accurate phenotyping techniques and integrate structural, physiological, and environmental information to simulate the overall understanding of how roots distribute in soil and interact with different environmental factors (Ndour *et al.*, 2017). It requires involvement of various fields, including plant physiology, soil science, engineering, mathematics, computer science, and so on. Many of these models have been developed and used to simulate the development of the plant with water acquisition and transportation (reviewed by Ndour *et al.*, 2017). These models aim to quantify RSA and set up descriptors of RSA, and eventually identify mechanisms of RSA and plasticity under given stress environments (reviewed by Ndour *et al.*, 2017). Recently, some new RSA models have been developed for various purposes to predict RSA and interaction with environments more accurately. This includes a root hydraulics model to estimate plant-scale hydraulic parameters from RSA (Meunier *et al.*, 2017), OPENSIMROOT for simulation of RSA, nutrient acquisition, and plant growth (Postma *et al.*, 2017), and RootBox to simulate matured RSA from seedling RSA (Leitner *et al.*, 2010; Zhao *et al.*, 2017). Challenges exist in developing and improving RSA models, such as the bottleneck of root phenotyping techniques, difficulties under field conditions, and integration of the plant growing ecosytems (reviewed by Ndour *et al.*, 2017). Overall, the future of RSA modeling relies on successful integration of all biological scales.

## Root system architecture in legumes and cereals

Roots are one of the major organs for transmitting different resources from soil, and thereby control productivity (Lynch, 1995). Plants have the capacity to customize their root architecture in order to adapt to different environments, including stress, through integrating the genetic programs controlling root growth (Jovanovic *et al.*, 2008). Plants with different genetic and nutritional conditions have shown extensive differences in root structure in terms of the number and distribution of root systems, which allows plants to recover efficiently when transiting from stress to normal conditions (López-Bucio *et al.*, 2003).

In the case of cereals, the root system is monocotyledonous, comprising primary root(s) generated from the seed embryo and axile nodal roots. The number of primary roots varies from one in warm-climate cereals to six or seven in cool-climate cereals. However, in the case of a dicotyledonous root system (Arabidopsis, *Medicago*, and legumes), a single primary root was generated from the embryo with successive orders of branch/lateral roots (Sorin *et al.*, 2005; Rich and Watt, 2013). The number and size of lateral roots are determining factors for RSA in legumes (Dubrovsky *et al.*, 2006). Comparative analysis of monocot (cereals and wheat) and dicot (legumes and lupin) root systems for length and water uptake suggested the existence of a longer root system in wheat compared with lupin. However, lupin had a higher water uptake per unit length as compared with wheat due to anatomical differences (Hamblin and Tennant, 1987; Bramley *et al.*, 2009). RSA in legumes is also partially determined by the symbiotic interaction along with soil conditions. Roots in legumes have N-fixing nodules for symbiotic interactions with soil bacteria (Oldroyd and Downie, 2004). A common regulatory pathway between nodule formation and root development was identified among legumes (Penmetsa *et al.*, 2003; Veereshlingam *et al.*, 2004; Bright *et al.*, 2005). RSA in symbiotic and non-symbiotic conditions is at least partially controlled by the same genes through an autoregulation mechanism controlling hypernodulating or supernodulating effects (Wopereis *et al.*, 2000; Krusell *et al.*, 2002).

## Genetic diversity and analysis of RSA in legumes

### *Soybean*

As a major cash crop, soybean has been intensively studied for RSA. Improved RSA has been shown to alleviate drought stress by increasing exploration for water and nutrients in soybean (Hoogenboom *et al.*, 1987). Natural variations in RSA were reported in soybean, and can be utilized for improvement of drought tolerance (Carter, 1989). In a field evaluation, a drought-tolerant landrace (PI 416937) showed more fibrous roots and exploration of a larger volume of soil (Hudak and Patterson, 1996). Recently several soybean accessions were identified showing promising RSA for extensive fibrous rooting, root length, or narrow root angle by screening of a core set (~400 lines) of the USDA Germplasm Collection. Genetic diversity in root anatomy of soybean is also reported, which affected water movement through root systems, as variations were observed in the number of metaxylems in soybean roots and the metaxylems were found to be induced during drought (Rincon *et al.*, 2003; Prince *et al.*, 2017). More metaxylems help in water transportation in the roots and protect yield under drought stress (Prince *et al.*, 2017).

QTL mapping has been conducted in soybean populations for RSA traits. Initially, five QTLs associated with fibrous rooting were mapped between the soybean lines Benning and PI 416937 under field conditions, with four out of the five QTLs donated by the drought-tolerant parent PI 416937 (Abdel-Haleem *et al.*, 2011). In a bi-parental recombinant inbred population, one QTL cluster was mapped for both root length and lateral root number with the donor alleles from an older cultivar, Dunbar (Manavalan *et al.*, 2015). This indicated the potential for some favorable alleles for RSA in some older soybean varieties and these genetic materials should be better candidates for breeding design (Table 1). Wild soybeans are valuable genetic resources for soybean improvement. One study identified four QTLs associated with surface area and root volume in a population of *Glycine max*×*Glycine soja*, and the favorable alleles of the QTLs were from the wild parent (PI 407162) (Prince *et al.*, 2015). Recent results show that one rare allele can promote lateral root growth with the potential to improve drought tolerance. A GWAS of 252 phenotypically diverse soybean germplasms pinpointed this allele, and this single nucleotide polymorphism (SNP) can cause more proliferative lateral rooting in soybean (unpublished results). Further screening of the USDA

**Table 1.** *Summary of QTLs associated with root system architecture and plasticity to drought in major legumes*

| Species | Trait | QTL name[a] | Donor parent[b] | Chro.[c] | Nearest marker[d] | $R^{2e}$ | Reference |
|---|---|---|---|---|---|---|---|
| Soybean | Fibrous rooting/ Surface area | FR_Gm01 | PI 416937 | 1 | Satt383 | 7.5 | Abdel-Haleem *et al.* (2011) |
| | | FR_Gm03 | PI 416937 | 3 | Satt339 | 13.5 | Abdel-Haleem *et al.* (2011) |
| | | FR_Gm04 | PI 416937 | 4 | Satt713 | 7.3 | Abdel-Haleem *et al.* (2011) |
| | | FR_Gm08 | PI 416937 | 8 | Satt228 | 12.8 | Abdel-Haleem *et al.* (2011) |
| | | FR_Gm20 | Benning | 20 | Sat_420 | 7.6 | Abdel-Haleem *et al.* (2011) |
| | | SA_Gm06 | PI 407162 | 6 | 4222.1.S1_10 | 13 | Prince *et al.* (2015) |
| | Root length | TRL_Gm06 | PI 407162 | 6 | 4222.1.S1_10 | 11 | Prince *et al.* (2015) |
| | | RDL3_Gm07 | PI 407162 | 7 | 8398.1.S1_11 | 10 | Prince *et al.* (2015) |
| | | TRL_Gm08 | Dunbar | 8 | Satt315 | 12.3 | Manavalan *et al.* (2015) |
| | Lateral root number | LRN_Gm08 | Dunbar | 8 | Satt315 | 11 | Manavalan *et al.* (2015) |
| | | GRL1 | PI 603713 | 16 | Gm16_29679156 | – | Unpublished data |
| | Root thickness | RDT2_Gm08 | PI 407162 | 7 | 59884.1.S1_8 | 15 | Prince *et al.* (2015) |
| Cowpea | Basal root angle | Brg10 | – | 10 | 4510_497 | – | Burridge *et al.* (2017) |
| | Root diameter | RD1 | – | 1 | 10905_418 | – | Burridge *et al.* (2017) |
| | Median width | MW6 | – | 6 | 2227_693 | – | Burridge *et al.* (2017) |
| | Width accumulation | WA10 | – | 10 | 4245_136 | – | Burridge *et al.* (2017) |
| Pea | Root length | rl1 | Messire | VI | OPZ17_1259 | 14 | Fondevilla *et al.* (2010, 2011) |
| | | rl2 | Messire | III | OPB11_1003 | 17 | Fondevilla *et al.* (2010, 2011) |
| | | rl3 | Messire | II | OPAI14_1353 | 11 | Fondevilla *et al.* (2010, 2011) |
| Common bean | Basal root angle | Brg1.1 | DOR364 | 1 | O20105G | 14 | Liao *et al.* (2004) |
| | | Brg5.1 | G19833 | 5 | K124G | 9.9 | Liao *et al.* (2004) |
| | | Brg5.2 | G19833 | 5 | GC13 | 15.9 | Liao *et al.* (2004) |
| Chickpea | Root length density | RLD4 | ICC 4958 | 4 | ICCM0249 | 12.1 | Jaganathan *et al.* (2015) |
| | Root surface area | RSA6 | ICC 4958 | 6 | H1F21 | 11 | Jaganathan *et al.* (2015) |
| | Root dry weight ratio | RDWR4 | ICC 4958 | 4 | ICCM0249 | 13.6 | Jaganathan *et al.* (2015) |
| | Rooting depth/root length density | QTL-hotspot | ICC 4958 | 4 | ICCM0249/ STMS11 | 11–47 | Jaganathan *et al.* (2015) |

[a]Name of the QTL that was generated in the corresponding reference or named afterwards in this review.
[b]Parental lines that contain the trait-favorable alleles.
[c]Chromosomes where the QTL are located.

Germplasm Collection found that the frequency of the favorable alleles at this locus is ~0.5% (unpublished results). As discussed above, several QTLs for RSA in soybean have been identified and validated (Table 1); however, the relationships between these QTLs and drought tolerance have not yet been evaluated. Therefore, further field evaluation of the effects of these QTLs on yield stability under drought conditions is needed.

### *Cowpea*

Cowpea [*Vigna unguiculata* (L.) Walp] is a primary protein source and food security crop in Africa and South America. Cowpea plays a significant role in producing nitrogen in low-input systems (Singh *et al.*, 1997, 2003; Huynh *et al.*, 2013). Cowpea is also cultivated to adapt environments commonly experiencing drought stress (Yadav *et al.*, 2015). Roots of 188 accession of cowpea were sampled from the field at the mature stage and analyzed for RSA (Bucksch *et al.*, 2014). Large variations in RSA were identified in this diverse set, including root length, diameters, branching angles, density, and spatial root distribution estimations. The accessions, such as SuVita-2 and UCR 3310, with promising RSA could be used to breed drought-tolerant cowpea varieties. QTL mapping work for RSA has been reported in cowpea recently. GWAS on the same diverse set of 188 accessions identified 18

QTL clusters from manually and digitally scored RSA traits (Burridge *et al.*, 2017). Subsequent comparisons of results from this root study with other field studies revealed four QTLs co-localized with seed weight per plant, pod number, or Striga (*Striga gesnerioides*) tolerance (listed in Table 1). These results suggest that selection for root phenotypes with MAS could be employed in breeding programs aiming to improve yield and yield stability in stress environments.

### Pea

Pea (*Pisum sativum* L.) is the most widely grown legume crop in Europe and the fourth most widely grown worldwide (FAO 2010). Pea is traditionally produced in marginal areas and has received little attention from producers compared with other legume crops, such as soybean. Evaluation of RSA in pea started in the early 2000s. Significant variations in RSA were observed in a core collection of 330 pea accessions, and the total root length and root dry weight were positively correlated with seed size (McPhee, 2005). The top 10% of lines for total root production were suggested in this report for a breeding program with PI 261631; an accession from Spain produced the greatest total root length, largest root/shoot weight ratio, and the largest root volume (McPhee, 2005). Initial QTL mapping was conducted in a recombinant inbred line (RIL) population derived from a cross between P665 and Messire. Three QTLs associated with root length were mapped on chromosomes (Chr.) 3 and 4 where they co-localized with QTLs for weed and fungal resistance (Fondevilla *et al.*, 2010, 2011; listed in Table 1). In the subsequent work, three QTLs were mapped for these drought tolerance-related traits and two of them were co-located with the previously reported root length QTL (Iglesias-García *et al.*, 2015). These results further confirmed the two QTLs associated with the RSA and revealed their real effects in drought tolerance.

### Common bean

Common bean (*Phaseolus vulgaris* L.) is the most important food legume, providing protein for a population of >500 million. It also provides necessary vitamins and micronutrients to over a billion people in developing countries (FAO, 2000). Common bean is usually grown in marginal areas, where soil is less fertile, and fertilizers and irrigation are not affordable. Therefore, improvement of stress resistance of common bean is urgent for these areas to maintain crop production. In an interspecific population, 86 RILs were derived from a cross between a deep-rooted genotype (DOR364) and a shallow-rooted genotype (G19833) (Liao *et al.*, 2004). QTL mapping was conducted in this population and 16 QTLs were mapped for root gravitropic traits, including basal root angle, shallow root length, and relative shallow basal root length, in which three basal root angle QTLs were found to be associated with deep rooting (Liao *et al.*, 2004; Table 1). The subsequent study identified that four QTLs associated with total root length in the field and one major QTL on Chr. B4 could explain 21% of phenotypic variation (Ochoa *et al.*, 2006; Table 1). In this study, adventitious root was positively correlated with shoot mass, and two major QTLs associated with adventitious root were also identified, together explaining 61% of phenotypic variation (Ochoa *et al.*, 2006). These results provide a potential genetic and molecular resource for MAS to develop new cultivars with improved RSA.

### Chickpea

Chickpea (*Cicer arietinum* L.) is the second most important food legume after common bean worldwide (FAOSTAT, 2014). It is mostly grown in low-input conditions especially in semi-arid regions of sub-Saharan Africa and South Asia, as an important component of subsistence farming. Drought stress alone accounts for 40% of yield losses annually to global chickpea production (Jukanti *et al.*, 2015). Strategic crop improvement programs rely on genetic improvement efforts aiming at incorporation of beneficial traits, such as better RSA, into elite cultivars (Subbarao *et al.*, 1995; Varshney *et al.*, 2009).

Root biomass and root length density that aid in greater soil moisture extraction were identified as important root traits during terminal drought (Kashiwagi *et al.*, 2006, 2007; Varshney *et al.*, 2011). Extensive phenotyping and genotyping of two intraspecific RIL populations [ICC 4958×ICC 1882 (ICCRIL03) and ICC 283×ICC 8261 (ICCRIL04)] were conducted to map QTLs for various drought tolerance traits, including RSA in chickpea (Varshney *et al.*, 2014). ICC 4958 and ICC 8261 were identified to be drought-tolerant genotypes with larger root systems (Saxena *et al.*, 1993; Kashiwagi *et al.*, 2005), and the other two contrasting parents, ICC 1882 and ICC 283, were landraces with shorter root systems (Kashiwagi *et al.*, 2005). Three QTLs, one each for root length density (CaLG04), root surface area (CaLG06), and root dry weight/total plant dry weight ratio (CaLG04), have been identified (Jaganathan *et al.*, 2015). This study also resulted in identification of an important genomic region, the '*QTL hotspot*', harboring QTLs (M-QTL and E-QTL) for several RSA and drought tolerance traits (Jaganathan *et al.*, 2015). Fine-mapping of this 'QTL-hotspot' delimited the candidate region from 29 cM to 14 cM on a genetic map, and several stress-related candidate genes were identified in this region (Jaganathan *et al.*, 2015). This '*QTL hotspot*' was introgressed from the donor parent ICC 4958 into JG 11 (a leading chickpea variety) using the marker-assisted backcrossing (MABC) strategy (Jaganathan *et al.*, 2015). Twenty-nine $BC_3F_2$ plants, referred to as introgression lines (ILs), showed greater rooting depth in all 29 ILs as compared with both recurrent parents (Jaganathan *et al.*, 2015). These ILs have shown better performance in comparison with the recurrent parents under rainfed as well as in irrigated environments (Jaganathan *et al.*, 2015). More recently, another study identified 13 QTLs associated with root traits in the same two intraspecific populations through the development of an SNP genotyping array for chickpea (Roorkiwal *et al.*, 2017).

### Lentil

Lentil (*Lens culinaris* Medik.) is considered as a staple food in developing countries for its rich source of proteins and micronutrients. It is considered as an important component in crop rotation programs due to its ability to improve soil fertility by atmospheric nitrogen fixation. Large genetic variability and high heritability in root and shoot traits related to drought tolerance were observed in 133 RILs from a cross of ILL6002 (superior root and shoot traits and grain yield)×ILL5888 (pure line, short cultivar with prostrate growth habit) (Idrissi *et al.*, 2015). Identification of QTLs or genes related to studied traits in this population would be a first step for starting MAS.

## Advances in breeding for drought tolerance

Lines with a deep root system are able to extract water from the soil and enable the plants to survive in water-limited conditions. Despite the importance of the root system, there have been limited efforts directed towards developing breeding lines with an improved root system. In the case of chickpea, the role of the root system in the improvement of crop productivity under water-limited conditions has been well studied and established. Greater exploitation of subsoil water due to deeper root systems has been shown to improve chickpea yield under drought conditions (Kashiwagi *et al.*, 2015). Similarly, in the case of soybean, the root system is important to protect yield under water-limited conditions. In order to determine the role of the root in crop productivity under water-limited conditions, different root traits including root length, root surface area, average root diameter, and average root volume were compared under normal and drought stress conditions (Zenis *et al.*, 2016). This study confirmed the importance of a better root system in drought tolerance.

Root traits have a direct impact on yield under drought stress among legumes (see table 2 in Farooq *et al.*, 2016), suggesting the root as a significant target for further breeding. Although there have been many efforts to identify genetic diversity and QTLs/genes that control RSA, there have been limited efforts to utilize these genetic resources in breeding. In chickpea, efforts have been undertaken to introgress the 'QTL-hotspot' region from line CaLG04 into the elite chickpea line (JG11) by the MABC strategy. Introgression lines have shown improved performance and enhanced yield under drought stress (Varshney *et al.*, 2013). Currently, efforts are being made to introgress this genomic region to several other elite chickpea lines (Pandey *et al.*, 2016). To date, rich genetic resources for RSA have been identified in legume crops, but those resources are yet to be implemented in breeding for developing stress-tolerant lines. Genotypes with differential RSA patterns should be utilized for developing stress-tolerant lines with improved RSA for each specific condition.

## Conclusions and future perspectives

The sustainability of crop production and food security is being threatened by the increasing unpredictability and severity of drought stress due to global climate changes. Genetic variations for stress tolerance in natural populations reflect the evolution of plants in diverse ecological conditions. Incorporation of these adapted natural genetic variations into breeding programs can enrich the current genetic diversity of stress tolerance and improve yield under stress. Screening germplasm for stress tolerance traits has provided donor resources with potential to improve yield under stress conditions. Identification of the genomic regions underlying these natural genetic variations and transferring the favorable alleles into elite germplasm with the assistance of genomic technologies will speed up the genetic improvement of stress tolerance in legumes. QTLs associated with RSA were identified in major legume crops, and associated DNA markers have been developed for MAS. Some of these QTLs or genes can be lost during domestication processes due to their linkage to negative loci or pleiotropic effects on yield. Subsequent fine-mapping is needed to determine whether the yield drag effects are due to pleiotropy or linkage to other negative loci. Screening a large population of segregants can break the tight linkage with the assistance of MAS. In addition, cloning and functional characterization of genes underlying the natural genetic variations can lead to understanding of the molecular and physiological pathways of stress adaptation, which will direct further improvement of stress tolerance in legumes.

High-throughput and cost-effective phenotyping and genotyping technologies are foundations of genomics-assisted breeding programs. High-throughput phenotyping platforms that monitor RSA are essential to advance our knowledge of drought tolerance. High-throughput phenotyping of RSA has focused on field-based approaches in crops such as maize (Trachsel *et al.*, 2011; York *et al.*, 2015), rice (Wissuwa *et al.*, 2016), and wheat (Richard *et al.*, 2015), with a recent shift to legume species, such as soybean, common bean, and cowpea (Manavalan *et al.*, 2015; Prince *et al.*, 2015; Burridge *et al.*, 2016). The adoption of high-throughput image analysis using DIRT will further improve the efficiency and accuracy of the phenotyping of RSA (Burridge *et al.*, 2016).

Until the last decade, legumes have fallen into the category of orphan crops with fewer genomic resources. Recent advances in sequencing technology have brought legumes into the category of genomic resource-rich crops. The availability of these resources resulted in the initiation of several crop improvement programs based on genomic-assisted breeding. MAS and genomic selection are the major genomic-assisted breeding approaches deployed in legume improvement programs (Pandey *et al.*, 2016). MAS is mostly used to address the issues related to simple traits that are controlled by one or two genes. However, for complex traits, MAS has not been that successful. Recently, genomic selection has emerged. It has the ability to target many complex traits at once and holds the potential for genetic improvement at a faster pace (Crossa *et al.*, 2017). High-precision phenotyping and genotyping have resulted in precise selection of genotypes with a reduced breeding cycle time by avoiding unnecessary rounds of phenotyping. The ability to capture both small and large effect genetic factors adds further advantages to the genomic selection strategy. Reduced cost of genotyping has resulted in establishment of a large range of genetic resources among legumes in the past decade. These resources are now providing required information for various genomic-assisted breeding approaches to enhance the genetic gains. Further establishment of high-throughput genotyping platforms in various legume crops has initiated the genomic selection efforts in legumes (Jain *et al.*, 2013).

Genetic modification has emerged as another important tool for crop improvement (Hussain, 2015). It provides an opportunity to edit targeted genes for specific needs. The targeted genome editing by the CRISPR/Cas9 (clustered regularly interspaced short palindromic repeat/CRISPR-associated protein 9) system (Jacobs *et al.*, 2015; Sun *et al.*, 2015) can enable further modification of RSA-related alleles to maximize the performance of the loci. In addition to soybean genome information, the release of reference genomes and continuous building up of resequencing information of various legume crops can strengthen the comparative legume genomics for discovery and characterization of key genes and gene families involved in stress tolerance. High-density genome variation information available for major legumes can facilitate haplotype mining of the traits of interest and can lead to the design of haplotype-assisted breeding. Integration of these genomic resources and technologies into next-generation breeding strategies will accelerate the genetic improvement of yield under stress conditions for legume crops.

## Acknowledgements

We acknowledge funding support from the United Soybean Board and the Agricultural Experiment Station Project, USDA-NIFA towards the soybean genomics and breeding research program at the University of Missouri.

## References

**Abate T, Alene AD, Bergvinson D, Shiferaw B, Silim S, Orr A, Asfaw S.** 2012. Tropical grain legumes in Africa and South Asia: knowledge and opportunities. Nairobi, Kenya: International Crops Research Institute for the Semi-Arid Tropics.

**Abdel-Haleem H, Lee GJ, Boerma RH.** 2011. Identification of QTL for increased fibrous roots in soybean. Theoretical and Applied Genetics **122,** 935–946.

**Addington RN, Donovan LA, Mitchell RJ, Vose JM, Pecot SD, Jack SB, Hacke UG, Sperry JS, Oren R.** 2006. Adjustments in hydraulic architecture of *Pinus palustris* maintain similar stomatal conductance in xeric and mesic habitats. Plant, Cell and Environment **29,** 535–545.

**Anderson W, Johansen C, Siddique KHM.** 2016. Addressing the yield gap in rainfed crops: a review. Agronomy for Sustainable Development **36,** 18.

**Andrews M, Hodge S.** 2010. Climate change, a challenge for cool season grain legume crop production. In: **Yadav S, Redden R**, eds. Climate change and management of cool season grain legume crops. Dordrecht: Springer, 1–10.

**Araki H, Morita S, Tatsumi J, Iijima M.** 2002. Physiomorphological analysis on axile root growth in upland rice. Plant Production Science **5,** 286–293.

**Babu RC, Nguyen BD, Chamarerk V, *et al*.** 2003. Genetic analysis of drought resistance in rice by molecular markers: association between secondary traits and field performance. Crop Science **43,** 1457–1469.

**Beebe S, Ramirez J, Jarvis A, Rao IM, Mosquera G, Bueno JM, Blair MW.** 2011. Genetic improvement of common beans and the challenges of climate change. In: **Yadav SS, Redden RJ, Hatfield JL, Lotze-Campen H, Hall AE**, ed. Crop adaptation to climate change. Chicheser, UK: John Wiley & Sons, Ltd.

**Bernier J, Kumar A, Spaner D, *et al*.** 2009. Characterization of the effect of rice drought tolerance qtl12.1 over a range of environments in the Philippines and eastern India. Euphytica **166,** 207–217.

**Bhatia VS, Singh P, Wani SP, Kesava Rao AVR, Srinivas K.** 2006. Yield gap analysis of soybean, groundnut, pigeonpea and chickpea in india using simulation modeling. Global Theme on Agroecosystems Report no. 31. Patancheru: International Crops Research Institute for the Semi-Arid Tropics.

**Blum A.** 2011. Plant breeding for water-limited environments. New York: Springer.

**Boyer JS.** 1983. Environmental stress and crop yields. In: **Raper CD, Kramer PJ**, eds. Crop reaction to water and temperature stresses in humid, temperate climates. Boulder, CO: Westview Press, 3–7.

**Bramley H, Turner NC, Turner DW, Tyerman SD.** 2009. Roles of morphology, anatomy, and aquaporins in determining contrasting hydraulic behavior of roots. Plant Physiology **150,** 348–364.

**Bright LJ, Liang Y, Mitchell DM, Harris JM.** 2005. The LATD gene of *Medicago truncatula* is required for both nodule and root development. Molecular Plant-Microbe Interactions **18,** 521–532.

**Bucksch A, Burridge J, York LM, Das A, Nord E, Weitz JS, Lynch JP.** 2014. Image-based high-throughput field phenotyping of crop roots. Plant Physiology **166,** 470–486.

**Burridge J, Jochuaa CN, Buckschc A, Lynch JP.** 2016. Legume shovelomics: high-throughput phenotyping of common bean (*Phaseolus vulgaris* L.) and cowpea (*Vigna unguiculata* subsp. *unguiculata*) root architecture in the field. Field Crops Research **192,** 21–32.

**Burridge JD, Schneider HM, Huynh BL, Roberts PA, Bucksch A, Lynch JP.** 2017. Genome-wide association mapping and agronomic impact of cowpea root architecture. Theoretical and Applied Genetics **130,** 419–431.

**Burton AL, Lynch JP, Brown KM.** 2013. Spatial distribution and phenotypic variation in root cortical aerenchyma of maize (*Zea mays* L.). Plant and Soil **367,** 263–274.

**Carter TE Jr**. 1989. Breeding for drought tolerance in soybean: where do we stand? In: **Pascale AJ**, ed. Proceedings of the World Soybean Conference IV. Buenos Aires, Argentina, 1001–1008.

**Chen L, Ren F, Zhou L, Wang QQ, Zhong H, Li XB.** 2012. The *Brassica napus* calcineurin B-Like 1/CBL-interacting protein kinase 6 (CBL1/CIPK6) component is involved in the plant response to abiotic stress and ABA signalling. Journal of Experimental Botany **63,** 6211–6222.

**Chen Y, Ghanem ME, Siddique KH.** 2017. Characterising root trait variability in chickpea (*Cicer arietinum* L.) germplasm. Journal of Experimental Botany **68,** 1987–1999.

**Christopher J, Christopher M, Jennings R, *et al*.** 2013. QTL for root angle and number in a population developed from bread wheats (*Triticum aestivum*) with contrasting adaptation to water-limited environments. Theoretical and Applied Genetics **126,** 1563–1574.

**Creelman RA, Mason HS, Bensen RJ, Boyer JS, Mullet JE.** 1990. Water deficit and abscisic acid cause differential inhibition of shoot *versus* root growth in soybean seedlings: analysis of growth, sugar accumulation, and gene expression. Plant Physiology **92,** 205–214.

**Crossa J, Pérez-Rodríguez P, Cuevas J, *et al*.** 2017. Genomic selection in plant breeding: methods, models, and perspectives. Trends in Plant Science **22,** 961–975.

**Daryanto S, Wang L, Jacinthe PA.** 2015. Global synthesis of drought effects on food legume production. PLoS One **10,** e0127401.

**Das A, Schneider H, Burridge J, Ascanio AK, Wojciechowski T, Topp CN, Lynch JP, Weitz JS, Bucksch A.** 2015. Digital imaging of root traits (DIRT): a high-throughput computing and collaboration platform for field-based root phenomics. Plant Methods **11,** 51.

**Dubrovsky JG, Gambetta GA, Hernández-Barrera A, Shishkova S, González I.** 2006. Lateral root initiation in Arabidopsis: developmental window, spatial patterning, density and predictability. Annals of Botany **97,** 903–915.

**FAO**. 2000. FAOSTAT database collections. Rome: Food and Agriculture Organization of the United Nations. URL: http://faostat.fao.org.

**FAO**. 2010. FAOSTAT database collections. Rome: Food and Agriculture Organization of the United Nations. URL: http://faostat.fao.org.

**FAOSTAT**. 2014. FAOSTAT database collections. Rome: Food and Agriculture Organization of the United Nations. URL: http://faostat.fao.org/site/567/default.aspx#ancor.

**Farooq M, Gogoi N, Barthakur S, Baroowa B, Bharadwaj N, Alghamdi SS, Siddique KHM.** 2016. Drought stress in grain legumes during reproduction and grain filling. Journal of Agronomy and Crop Science **203,** 81–102.

**Fenta BA, Beebe SE, Kunert KJ, Burridge JD, Barlow KM, Lynch PJ.** 2014. Field phenotyping of soybean roots for drought stress tolerance. Agronomy **4,** 418–435.

**Fleury D, Jefferies S, Kuchel H, Langridge P.** 2010. Genetic and genomic tools to improve drought tolerance in wheat. Journal of Experimental Botany **61,** 3211–3222.

**Fondevilla S, Almeida NF, Satovic Z, Rubiales D, Vaz-Patto MC, Cubero JI, Torres AM.** 2011. Identification of common genomic regions controlling resistance to *Mycosphaerella pinodes*, earliness and architectural traits in different pea genetic backgrounds. Euphytica **182,** 43–52.

**Fondevilla S, Fernández-Aparicio M, Satovic Z, *et al*.** 2010. Identification of quantitative trait loci for specific mechanisms of resistance to *Orobanche crenata* Forsk in pea (*Pisum sativum* L.). Molecular Breeding **25,** 259–272.

**Forster BP, Thomas WTB, Chloupek O.** 2005. Genetic controls of barley root systems and their associations with plant performance. Aspects of Applied Biology **73,** 199–204.

**Foyer CH, Lam HM, Nguyen HT, *et al*.** 2016. Neglecting legumes has compromised human health and sustainable food production. Nature Plants **2,** 16112.

**Garay AF, Wilhelm WW.** 1983. Root system characteristics of two soybean isolines undergoing water stress conditions. Agronomy Journal **75,** 973–977.

**Gaur PM, Krishnamurthy L, Kashiwagi J.** 2008. Improving drought-avoidance root traits in chickpea (*Cicer arietinum* L.)—current status of research at ICRISAT. Plant Production Science **11,** 3–11.

**Giuliani S, Sanguineti MC, Tuberosa R, Bellotti M, Salvi S, Landi P.** 2005. Root-ABA1, a major constitutive QTL, affects maize root architecture and leaf ABA concentration at different water regimes. Journal of Experimental Botany **56,** 3061–3070.

**Hamblin AP, Tennant D.** 1987. Root length density and water uptake in cereals and grain legumes: how well are they correlated. Australian Journal of Agricultural Research **38,** 513–527.

**Hammer G, Dong Z, Mclean G, Doherty A, Messina C, Schussler J, Zinselmeier C, Paszkiewicz S, Cooper M.** 2009. Can changes in canopy and/or root system architecture explain historical maize yield trends in the U.S. Corn Belt? Crop Science **49,** 299–312.

**Hatfield JL, Boote KJ, Kimball BA, Ziska LH, Izaurralde RC, Ort D, Thomson AM, Wolfe D.** 2011. Climate impacts on agriculture: implications for crop production. Agronomy Journal **103,** 351–370.

**Hoogenboom G, Huck MG, Peterson CM.** 1987. Root growth rate of soybean as affected by drought stress. Agronomy Journal **79,** 607–614.

**Hudak CM, Patterson RP.** 1995. Vegetative growth analysis of a drought resistant soybean plant introduction. Crop Science **35,** 464–471.

**Hudak CM, Patterson RP.** 1996. Root distribution and soil moisture depletion pattern of a drought-resistant soybean plant introduction. Agronomy Journal **88,** 478–485.

**Hund A, Reimer R, Messmer R.** 2011. A consensus map of QTLs controlling the root length of maize. Plant and Soil **344,** 143–158.

**Hussain B.** 2015. Modernization in plant breeding approaches for improving biotic stress resistance in crop plants. Turkish Journal of Agriculture and Forestry **39,** 515–530.

**Huynh B, Close TJ, Roberts PA, *et al*.** 2013. Gene pools and the genetic architecture of domesticated cowpea. Plant Genome **6,** 1–8.

**Idrissi O, Houasli CH, Udupa SM, De Keyser E, Van Damme P, De Riek J.** 2015. Genetic variability for root and shoot traits in a lentil (*Lens culinaris* Medik.) recombinant inbred line population and their association with drought tolerance. Euphytica **204,** 693–709.

**Iglesias-García R, Prats E, Fondevilla S, Satovic Z, Rubiales D.** 2015. Quantitative trait loci associated to drought adaptation in pea (*Pisum sativum* L.). Plant Molecular Biology Reporter **33,** 1768–1778.

**Jacobs TB, LaFayette PR, Schmitz RJ, Parrott WA.** 2015. Targeted genome modifications in soybean with CRISPR/Cas9. BMC Biotechnology **15,** 16.

**Jaganathan D, Thudi M, Kale S, Azam S, Roorkiwal M, Gaur PM, Kishor PB, Nguyen H, Sutton T, Varshney RK.** 2015. Genotyping-by-sequencing based intra-specific genetic map refines a ‘QTL-hotspot’ region for drought tolerance in chickpea. Molecular Genetics and Genomics **290,** 559–571.

**Jain M, Misra G, Patel RK, *et al*.** 2013. A draft genome sequence of the pulse crop chickpea (*Cicer arietinum* L.). The Plant Journal **74,** 715–729.

**Jovanovic M, lefebvre V, Laporte P, Gonzalez-rizzo S, Lelandais-briere C, Frugier F, Hartmann C, Crespi M.** 2008. How the environment regulates root architecture in dicots. Advances in Botanical Research **46,** 35–74.

**Jukanti AK, Bhatt R, Sharma R, Kalia RK.** 2015. Morphological, agronomic, and yield characterization of cluster bean (*Cyamopsis tetragonoloba* L.) germplasm accessions. Journal of Crop Science and Biotechnology **18,** 83–88.

**Kashiwagi J, Krishnamurthy L, Crouch JH, Serraj R.** 2006. Variability of root length density and its contributions to seed yield in chickpea (*Cicer arietinum* L.) under terminal drought stress. Field Crops Research **95,** 171–181.

**Kashiwagi J, Krishnamurthy L, Purushothaman R, Upadhyaya HD, Gaur PM, Gowda CLL, Ito O, Varshney RK.** 2015. Scope for improvement of yield under drought through the root traits in chickpea (*Cicer arietinum* L.). Field Crops Research **170,** 47–54.

**Kashiwagi J, Krishnamurthy L, Serraj R, Crouch JH, Panwar S.** 2007. Rapid and reliable screening technique for root traits in chickpea. Journal of Food Legumes **20,** 67–70.

**Kashiwagi J, Krishnamurthy L, Upadhyaya HD, Krishna H, Chandra S, Vadez V, Serraj R.** 2005. Genetic variability of drought-avoidance root traits in the mini-core germplasm collection of chickpea (*Cicer arietinum* L.). Euphytica **146,** 213–222.

**Kato Y, Abe J, Kamoshita A, Yamagishi J.** 2006. Genotypic variation in root growth angle in rice (*Oryza sativa* L.) and its association with deep root development in upland fields with different water regimes. Plant and Soil **287,** 117–129.

**Kramer PJ.** 1969. Plant and soil water relationships: a modern synthesis. New York: McGrawHill.

**Krusell L, Madsen LH, Sato S, *et al*.** 2002. Shoot control of root development and nodulation is mediated by a receptor-like kinase. Nature **420,** 422–426.

**Landi P, Giuliani S, Salvi S, Ferri M, Tuberosa R, Sanguineti MC.** 2010. Characterization of root-yield-1.06, a major constitutive QTL for root and agronomic traits in maize across water regimes. Journal of Experimental Botany **61,** 3553–3562.

**Leitner D, Klepsch S, Bodner G, Schnepf A.** 2010. A dynamic root system growth model based on L-Systems. Plant and Soil **332,** 177–192.

**Lesk C, Rowhani P, Ramankutty N.** 2016. Influence of extreme weather disasters on global crop production. Nature **529,** 84–87.

**Levitt J.** 1972. Responses of plants to environmental stresses. New York: Academic Press.

**Liao H, Yan X, Rubio G, Beebe SE, Blair MW, Lynch JP.** 2004. Genetic mapping of basal root gravitropism and phosphorus acquisition efficiency in common bean. Functional Plant Biology **31,** 959–970.

**Lobell DB, Roberts MJ, Schlenker W, Braun N, Little BB, Rejesus RM, Hammer GL.** 2014. Greater sensitivity to drought accompanies maize yield increase in the U.S. Midwest. Science **344,** 516–519.

**López-Bucio J, Cruz-Ramírez A, Herrera-Estrella L.** 2003. The role of nutrient availability in regulating root architecture. Current Opinion in Plant Biology **6,** 280–287.

**Lynch J.** 1995. Root architecture and plant productivity. Plant Physiology **109,** 7–13.

**Lynch JP.** 2011. Root phenes for enhanced soil exploration and phosphorus acquisition: tools for future crops. Plant Physiology **156,** 1041–1049.

**Mace ES, Singh V, Van Oosterom EJ, Hammer GL, Hunt CH, Jordan DR.** 2012. QTL for nodal root angle in sorghum (*Sorghum bicolor* L. Moench) co-locate with QTL for traits associated with drought adaptation. Theoretical and Applied Genetics **124,** 97–109.

**MacMillan K, Emrich K, Piepho HP, Mullins CE, Price AH.** 2006. Assessing the importance of genotype × environmental interaction for root traits in rice using a mapping population II: conventional QTL analysis. Theoretical and Applied Genetics **113,** 953–964.

**Manavalan LP, Prince SJ, Musket TA, *et al*.** 2015. Identification of novel QTL governing root architectural traits in an interspecific soybean population. PLoS One **10,** e0120490.

**Manschadi AM, Christopher J, Devoil P, Hammer GL.** 2006. The role of root architectural traits in adaptation of wheat to water-limited environments. Functional Plant Biology **33,** 823–837.

**Maseda PH, Fernández RJ.** 2006. Stay wet or else: three ways in which plants can adjust hydraulically to their environment. Journal of Experimental Botany **57,** 3963–3977.

**McPhee K.** 2005. Variation for seedling root architecture in the core collection of pea germplasm. Crop Science **45,** 1758–1763.

**Meister R, Rajani MS, Ruzicka D, Schachtman DP.** 2014. Challenges of modifying root traits in crops for agriculture. Trends in Plant Science **19,** 779–788.

**Meunier F, Couvreur V, Draye X, Vanderborght J, Javaux M.** 2017. Towards quantitative root hydraulic phenotyping: novel mathematical functions to calculate plant-scale hydraulic parameters from root system functional and structural traits. Journal of Mathematical Biology **75,** 1133–1170.

**Mickelbart MV, Hasegawa PM, Bailey-Serres J.** 2015. Genetic mechanisms of abiotic stress tolerance that translate to crop yield stability. Nature Reviews. Genetics **16,** 237–251.

**Mourtzinis S, Specht JE, Lindsey LE, *et al*.** 2015. Climate-induced reduction in US-wide soybean yields underpinned by region- and in-season-specific responses. Nature Plants **1,** 14026.

**Ndour A, Vadez V, Pradal C, Lucas M.** 2017. Virtual plants need water too: functional–structural root system models in the context of drought tolerance breeding. Frontiers in Plant Science **8,** 1577.

**Newman PR, Moser LE.** 1988. Grass seedling emergence, morphology, and establishment as affected by planting depth. Agronomy Journal **80,** 383–387.

**Nguyen H, Babu RC, Blum A.** 1997. Breeding for drought resistance in rice: physiology and molecular genetics considerations. Crop Science **37,** 1426–1434.

**Nielsen KL, Lynch JP, Weiss HN.** 1997. Fractal geometry of bean root systems: correlations between spatial and fractal dimension. American Journal of Botany **84,** 26–33.

**Ochoa I, Blair M, Lynch J.** 2006. QTL analysis of adventitious root formation in common bean under contrasting phosphorus availability. Crop Science **46,** 1609–1621.

**Oldroyd GE, Downie JA.** 2004. Calcium, kinases and nodulation signalling in legumes. Nature Reviews. Molecular Cell Biology **5,** 566–576.

**Osmont KS, Sibout R, Hardtke CS.** 2007. Hidden branches: developments in root system architecture. Annual Review of Plant Biology **58,** 93–113.

**Paez-Garcia A, Motes CM, Scheible WR, Chen R, Blancaflor EB, Monteros MJ.** 2015. Root traits and phenotyping strategies for plant improvement. Plants **4,** 334–355.

**Pandey MK, Roorkiwal M, Singh VK, Ramalingam A, Kudapa H, Thudi M, Chitikineni A, Rathore A, Varshney RK.** 2016. Emerging genomic tools for legume breeding: current status and future prospects. Frontiers in Plant Science **7,** 455.

**Pantalone VR, Rebetzke GJ, Burton JW, Carter TE.** 1996. Phenotypic evaluation of root traits in soybean and applicability to plant breeding. Crop Science **36,** 456–459.

**Passioura JB.** 1983. Root and drought resistance. Agricultural Water Management **7,** 265–280.

**Passioura JB.** 1977. Grain yield, harvest index and water use of wheat. Journal of the Australian Institute of Agricultural Science **43,** 117–120.

**Penmetsa RV, Frugoli JA, Smith LS, Long SR, Cook DR.** 2003. Dual genetic pathways controlling nodule number in *Medicago truncatula*. Plant Physiology **131,** 998–1008.

**Postma JA, Kuppe C, Owen MR, Mellor N, Griffiths M, Bennett MJ, Lynch JP, Watt M.** 2017. OpenSimRoot: widening the scope and application of root architectural models. New Phytologist **215,** 1274–1286.

**Prince SJ, Murphy M, Mutava RN, Durnell LA, Valliyodan B, Shannon JG, Nguyen HT.** 2017. Root xylem plasticity to improve water use and yield in water-stressed soybean. Journal of Experimental Botany **68,** 2027–2036.

**Prince SJ, Song L, Qiu D, *et al*.** 2015. Genetic variants in root architecture-related genes in a *Glycine soja* accession, a potential resource to improve cultivated soybean. BMC Genomics **16,** 132.

**Purushothaman R, Mainassara ZA, Nalini M, Rajaram P, Krishnamurthy L, Cholenahalli LG.** 2013. Root anatomical traits and their possible contribution to drought tolerance in grain legumes. Plant Production Science **16,** 1–8.

**Rich SM, Watt M.** 2013. Soil conditions and cereal root system architecture, review and considerations linking Darwin and Weaver. Journal of Experimental Botany **64,** 1193–1208.

**Richard CA, Hickey LT, Fletcher S, Jennings R, Chenu K, Christopher JT.** 2015. High-throughput phenotyping of seminal root traits in wheat. Plant Methods **11,** 13.

**Rincon CA, Raper CD, Patterson RP Jr**. 2003. Genotypic differences in root anatomy affecting water movement through roots of soybean. International Journal of Plant Science **164,** 543–551.

**Roorkiwal M, Jain A, Kale SM, Doddamani D, Chitikineni A, Thudi M, Varshney RK.** 2017. Development and evaluation of high density SNP array (Axiom® CicerSNP Array) for high resolution genetic mapping and breeding applications in chickpea. Plant Biotechnology Journal doi: 10.1111/pbi.12836.

**Rosenzweig C, Elliott J, Deryng D, *et al*.** 2014. Assessing agricultural risks of climate change in the 21st century in a global gridded crop model intercomparison. Proceedings of the National Academy of Sciences, USA **111,** 3268–3273.

**Sadok W, Sinclair TR.** 2011. Crops yield increase under water–limited conditions: review of recent physiological advances for soybean genetic improvement. Advances in Agronomy **113,** 313–337.

**Salazar-Henao JE, Vélez-Bermúdez IC, Schmidt W.** 2016. The regulation and plasticity of root hair patterning and morphogenesis. Development **143,** 1848–1858.

**Saxena NP, Krishnamurthy L, Johansen C.** 1993. Registration of a drought-resistant chickpea germplasm. Crop Science **33,** 1424.

**Sayar R, Khemira H, Kharrat M.** 2007. Inheritance of deeper root length and grain yield in half-diallel durum wheat (*Triticum durum*) crosses. Annals of Applied Biology **151,** 213–220.

**Singh BB, Ajeigbe HA, Tarawali SA, Fernández-Rivera S, Abubakar M.** 2003. Improving the production and utilization of cowpea as food and fodder. Field Crops Research **84,** 169–177.

**Singh BB, Chambliss OL, Sharma B.** 1997. Recent advances in cowpea breeding. In: **Singh BB, Mohan-Raj DR, Dashiell KE, Jackai LEN**, eds. Advances in cowpea research. Ibadan, Nigeria: The International Institute of Tropical Agriculture (IITA) and Iberaki, Japan: Japan International Research Center for Agricultural Sciences, 30–49.

**Singh V, van Oosterom EJ, Jordan DR, Hunt CH, Hammer GL.** 2011. Genetic variability and control of nodal root angle in sorghum. Crop Science **51,** 2011–2020.

**Sorin C, Bussell JD, Camus I, *et al*.** 2005. Auxin and light control of adventitious rooting in Arabidopsis require ARGONAUTE1. The Plant Cell **17,** 1343–1359.

**Sponchiado BN, White JW, Castillo JA, Jones PG.** 1989. Root growth of four common bean cultivars in relation to drought tolerance in environments with contrasting soil types. Experimental Agriculture **25,** 249–257.

**Steele KA, Price AH, Shashidhar HE, Witcombe JR.** 2006. Marker-assisted selection to introgress rice QTLs controlling root traits into an Indian upland rice variety. Theoretical and Applied Genetics **112,** 208–221.

**Subbarao GV, Johansen C, Slinkard AE, Nageswara Rao RC, Saxena NP, Chauhan YS, Lawn RJ.** 1995. Strategies for improving drought resistance in grain legumes. Critical Reviews in Plant Sciences **14,** 469–523.

**Sun X, Hu Z, Chen R, Jiang Q, Song G, Zhang H, Xi Y.** 2015. Targeted mutagenesis in soybean using the CRISPR–Cas9 system. Scientific Reports **5,** 10342.

**Takeda S, Matsuoka M.** 2008. Genetic approaches to crop improvement: responding to environmental and population changes. Nature Reviews. Genetics **9,** 444–457.

**Trachsel S, Kaeppler SM, Brown KM, Lynch JP.** 2011. Shovelomics: high throughput phenotyping of maize (*Zea mays* L.) root architecture in the field. Plant and Soil **341,** 75–87.

**Tuberosa R, Salvi S, Sanguineti MC, Landi P, Maccaferri M, Conti S.** 2002. Mapping QTLs regulating morpho-physiological traits and yield: case studies, shortcomings and perspectives in drought-stressed maize. Annals of Botany **89,** 941–963.

**Turner NC.** 2000. Drought resistance: a comparison of two frameworks. In: **Saxena NP, Johansen C, Chauhan YS, Rao RCN**, eds. Management of agriculture and drought: agronomic and genetic options. New Delhi, India: Oxford University Press, 89–102.

**Turner NC, Begg JE, Tonnet ML.** 1978. Osmotic adjustment of sorghum and sunflower crops in response to water deficits and its influence on the water potential at which stomata close. Australian Journal of Plant Physiology **5,** 597–608.

**Turner NC, Wright GC, Siddique KHM.** 2001. Adaptation of grain legumes (pulses) to water-limited environments. Advances in Agronomy **71,** 193–231.

**Uga Y, Sugimoto K, Ogawa S, *et al*.** 2013. Control of root system architecture by DEEPER ROOTING 1 increases rice yield under drought conditions. Nature Genetics **45,** 1097–1102.

**Varshney RK, Gaur PM, Chamarthi SK, *et al*.** 2013. Fast-track introgression of 'QTL-hotspot' for root traits and other drought tolerance traits in JG 11, an elite and leading variety of chickpea. Plant Genome **6,** doi: 10.3835/plantgenome2013.07.0022.

**Varshney RK, Hiremath PJ, Lekha P, *et al*.** 2009. A comprehensive resource of drought- and salinity-responsive ESTs for gene discovery and marker development in chickpea (*Cicer arietinum* L.). BMC Genomics **10,** 523.

**Varshney RK, Mir RR, Bhatia S, *et al*.** 2014. Integrated physical, genetic and genome map of chickpea (*Cicer arietinum* L.). Functional and Integrative Genomics **14,** 59–73.

**Varshney RK, Pazhamala L, Kashiwagi J, Gaur PM, Krishnamurthy L, Hoisington D.** 2011. Genomics and physiological approaches for root trait breeding to improve drought tolerance in chickpea (*Cicer arietinum* L.). In: **Costa de Oliveira A, Varshney RK**, eds. Root genomics. Dordrecht, The Netherlands: Springer, 233–250.

**Varshney RK, Singh VK, Hickey JM, *et al*.** 2015. Analytical and decision support tools for genomics-assisted breeding. Trends Plant Science **15,** S1360–S1385.

**Veereshlingam H, Haynes JG, Penmetsa RV, Cook DR, Sherrier DJ, Dickstein R.** 2004. *nip*, a symbiotic *Medicago truncatula* mutant that forms root nodules with aberrant infection threads and plant defense-like response. Plant Physiology **136,** 3692–3702.

**Wasson AP, Richards RA, Chatrath R, Misra SC, Prasad SV, Rebetzke GJ, Kirkegaard JA, Christopher J, Watt M.** 2012. Traits and selection strategies to improve root systems and water uptake in water-limited wheat crops. Journal of Experimental Botany **63,** 3485–3498.

**Wissuwa M, Kretzschmar T, Rose TJ.** 2016. From promise to application: root traits for enhanced nutrient capture in rice breeding. Journal of Experimental Botany **67,** 3605–3615.

**Wopereis J, Pajuelo E, Dazzo FB, Jiang Q, Gresshoff PM, De Bruijn FJ, Stougaard J, Szczyglowski K.** 2000. Short root mutant of *Lotus japonicus* with a dramatically altered symbiotic phenotype. The Plant Journal **23,** 97–114.

**Wu Y, Spollen WG, Sharp RE, *et al*.** 1994. Root growth maintenance at low water potentials. Plant Physiology **106,** 607–615.

**Yadav N, Kaur D, Malviya R, Rathore BS.** 2015. Evaluation of the nutritional and antioxidant properties of selected cowpea (*Vigna unguiculata*) cultivars. International Journal of Food and Nutritional Sciences **4,** 2320–7876.

**York LM, Galindo-Castañeda T, Schussler JR, Lynch JP.** 2015. Evolution of US maize (*Zea mays* L.) root architectural and anatomical phenes over the past 100 years corresponds to increased tolerance of nitrogen stress. Journal of Experimental Botany **66,** 2347–2358.

**York LM, Lynch JP.** 2015. Intensive field phenotyping of maize (*Zea mays* L.) root crowns identifies phenes and phene integration associated with plant growth and nitrogen acquisition. Journal of Experimental Botany **66,** 5493–5505.

**Yu LX, Ray JD, O'Toole JC, Nguyen HT.** 1995. Use of wax–petrolatum layers for screening rice root penetration. Crop Science **35,** 684–687.

**Zenis A, Kantartzi S, Meksem K, Kassem MA.** 2016. Influence of drought stress on several root traits and their correlation with seed protein and oil contents in soybean. Atlas Journal of Biology **2016,** 267–273.

**Zhao J, Bodner G, Rewald B, Leitner D, Nagel KA, Nakhforoosh A.** 2017. Root architecture simulation improves the inference from seedling root phenotyping towards mature root systems. Journal of Experimental Botany **68,** 965–982.

**Zheng HG, Babu RC, Pathan MS,Ali L, Huang N, Courtois B, Nguyen T.** 2000. Quantitative trait loci for root-penetration ability and root thickness in rice: comparison of genetic backgrounds. Genome **43,** 53–61.