The hidden half comes into the spotlight: Peeking inside the black box of root developmental phases

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ABSTRACT
Efficient use of natural resources (e.g., light, water, and nutrients) can be improved with a tailored developmental program that maximizes the lifetime and fitness of plants. In plant shoots, a developmental phase represents a time window in which the meristem triggers the development of unique morphological and physiological traits, leading to the emergence of leaves, flowers, and fruits. Whereas developmental phases in plant shoots have been shown to enhance food production in crops, this phenomenon has remained poorly investigated in roots. In light of recent advances, we suggest that root development occurs in three main phases: root apical meristem appearance, foraging, and senescence. We provide compelling evidence suggesting that these phases are regulated by at least four developmental pathways: autonomous, non-autonomous, hormonal, and periodic. Root developmental pathways differentially coordinate organ plasticity, promoting morphological alterations, tissue regeneration, and cell death regulation. Furthermore, we suggest how nutritional checkpoints may allow progression through the developmental phases, thus completing the root life cycle. These insights highlight novel and exciting advances in root biology that may help maximize the productivity of crops through more sustainable agriculture and the reduced use of chemical fertilizers.

Key words: developmental transitions, plasticity, cell fates, nutritional checkpoints, root development, root clock

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ROOTS: AMAZING ORGANS WITH A UNIQUE DEVELOPMENTAL CLOCK
The developmental clock describes a set of timekeepers that define the emergence of cells, tissues, and organs according to time. This clock defines the unique genetic, biochemical, and morphological features of the organism. Its complexity varies widely between or even within species and depends on embryonic patterns. Embryogenesis encompasses the formation of embryos and the completion of asymmetric cell divisions. Postembryonic development is the process whereby an organism builds its body, forming new tissues and organs. Postembryonic development of entire organs is prominent in plants but is much rarer in animals. Because plants are sessile organisms, the uncoupling of embryos and organogenesis is crucial for plant survival in different environments. Accordingly, plants do not necessarily pre-establish the number and size of organs during embryogenesis; instead, both postembryonic development and developmental phases ensure that plants thrive under diverse environmental conditions.

In the last decades, factors that control plant shoot development have been identified, and three developmental phase transitions have been recognized: seed germination-to-juvenile vegetative growth, the juvenile vegetative-to-adult vegetative phase, and the adult vegetative-to-reproductive phase (Poethig, 1990, 2003; Baurle and Dean, 2006; Huijser and Schmid, 2011; Yu et al., 2015a). Seed germination denotes the embryo-to-postembryonic transition. This is followed by juvenile vegetative growth, during which the plant remains unable to flower. Finally, during the floral transition, the plant enters the reproductive phase, whose highlight is the occurrence of meiosis during gametogenesis.

The transcription factor SHORT VEGETATIVE PHASE (SVP), known to delay floral transition, has recently been suggested to negatively regulate cambial cell proliferation in roots and increase total xylem vessel number in Arabidopsis thaliana (Arabidopsis) (Zhang et al., 2019a). Furthermore, the circadian clock component EARLY FLOWERING 4 (ELF4) was demonstrated to move from shoots to roots, delivering circadian cues in a temperature-dependent manner in Arabidopsis (Chen et al.,...
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2020). Thus, the proteins and temperatures that control phase transition in shoots seem also to regulate root development and synchronize the two organs. The root apical meristem (RAM) undergoes self-renewal and continuous differentiation as it emerges from the meristematic zone. Programmed cell death (PCD) in the root cap promotes lateral root spacing (Möller et al., 2017; Motte et al., 2019) and shifts the cell arrangement in the plant roots, possibly contributing to the regulation of developmental phases.

The first layer of root cap cells was recently shown to be characterized by cell-wall modifications resembling those seen in the cuticle of aerial plant organs. This cuticle is essential for root protection during seedling establishment and for lateral root formation in Arabidopsis (Berhin et al., 2019). Suberin, a hydrophobic biopolymer with a long chain formed by the binding of fatty acids and glycerol, may be deposited on the cell walls of specific root parts, including the endodermis, in response to environmental conditions in a process called suberization. Although suberization also occurs in other root parts (Watanabe et al., 2013), nutritional changes can promote endodermal differentiation dictated by the suberization status of this cell layer, revealing the functional and anatomical plasticity of adult roots in Arabidopsis (Barberon et al., 2016).

Unlike in animals, postembryonic development in plants relies on the asymmetric orientation of cell-division planes, which determines specific cell fates. Thus, we posit that unique developmental features are likely to characterize different plant species. Root stem cells may be reconstituted from multiple cell types, whereby a rapid transition in cell identity precedes de novo stem cell formation and follows the same sequence seen during embryonic root formation in Arabidopsis (Efroni et al., 2016). This indicates that the regenerative potential of the root primordium is comparable to that of the shoot apical meristem (SAM) (Rosspopoff et al., 2017). A cell’s relative position in the root is closely associated with its identity but may change during regeneration or restoration of tissue after drastic damage (Efroni, 2018). The existing concept of cell differentiation has been the target of criticism within the plant biology community, as a more fluid, flexible view of identity during cell transitions and differentiation is emerging (Sugimoto et al., 2011; Ikeuchi et al., 2016; Efroni, 2018). The identity acquired by distinct root cell types appears to be mediated by the stele-derived signaling peptide Casparian strip integrity factor 2 (CIF2), which stabilizes the transcription factor SHORT-ROOT (SHR) and promotes cell differentiation in the endodermis of Arabidopsis (Drapek et al., 2018). Studies on cell differentiation in young roots that display a low level of branching have excluded the influence of root cell ontogeny on developmental time. Quiescent center (QC) cells in the RAM are associated with spatial and developmental time status, and distance from the QC is closely linked to cell-differentiation potential in Arabidopsis (Wendrich et al., 2017). Accordingly, the QC proximal region produces two opposing gradients for transcriptional regulation, differentiation, and stemness (i.e., the ability of a cell to resume self-renewal), causing the expression of gradient-related genes to vary with root zone ontogeny (Wendrich et al., 2017). Collectively, the specific cell fates in the RAM appear to dictate developmental decisions, hinting at distinct developmental phases for roots.

Numerous studies have described developmental phase transitions in shoots (Huijser and Schmid, 2011; Yu et al., 2015a) but not in roots. We posit the existence of at least three distinct developmental phases in the roots. First is the RAM appearance phase, which is characterized by higher rates of cell proliferation than of differentiation. The root-foraging phase that follows contains more differentiated than undifferentiated cells, together with higher nutrient uptake from the rhizosphere. Finally, the root-senescence phase exhibits a constant decline and, latterly, an absence of cell proliferation/differentiation and nutrient uptake but increasing rates of cell death. Here, we provide new insights into root biology, which may assist in the development of next-generation crops that exhibit tailored root growth as well as improved uptake and use of nutrients to meet crop requirements. Accordingly, we posit that a late root-foraging phase may improve nitrogen (N) uptake and utilization, whereas early branching may improve the efficiency of phosphorus (P) utilization.

ROOT DEVELOPMENTAL PHASES

Roots exhibit diverse forms and functions: they include fine, pioneer, coarse, absorptive, and transport roots (McCormack et al., 2015). A growing body of evidence suggests that their lifespan depends on the RAM stage, akin to regulation of the SAM in shoots (Poethig, 2003; Baurle and Dean, 2006). Root developmental phases reflect cell fates associated with the RAM, regardless of the species or root class, and affect the entire root. This is likely to be similar to the situation described for shoot developmental phases, which display relatively little differentiation across flowering plant species (Pujar et al., 2006). Establishment of the QC promotes the maintenance and function of the whole root system, as may be observed in Arabidopsis and cereals, including rice (Oryza sativa), barley (Hordeum vulgare), and maize (Zea mays) (Strotmann and Stahl, 2021). Although cereals are likely to harbor more complex root systems than Arabidopsis (Strotmann and Stahl, 2021), these results indicate that the QC in the RAM seems to modulate all root developmental phases.

RAM appearance phase

Following gamete fusion, embryos undergo asymmetric cell division. During early development, distinct hormonal response zones form in the Arabidopsis embryo, establishing stable cell-division patterns and inducing the growth of the root vasculature, such that a functional phloem exists prior to seed germination (De Rybel et al., 2014a, 2014b). In response to adequate stimuli, seeds germinate, and the SAM and RAM give rise to the plant’s aerial and terrestrial structures, respectively (Figure 1). Importantly, during the RAM appearance phase, the RAM utilizes seed reserves to sustain cell division.

The BIRD family of transcription factors organizes ground tissue during embryogenesis. BIRD and SCARECROW (SCR) transcription factors regulate postembryonic growth in roots, defining stem cell zones, cell divisions, and cell layers in Arabidopsis (Moreno-Risueno et al., 2015). Regardless of SHR and SCR, root epidermis identity is limited by SCHIZORIZA (SCZ), which suppresses subepidermal cell fates and root hair formation in...
Arabidopsis (Mylona et al., 2002). By contrast, ARABIDOPSIS CRINKLY 4 (ACR4) has direct implications for intercellular communication and is required for the formation of epidermis-related tissues in the outer layers (Tanaka et al., 2002; Watanabe et al., 2004). The acr4 mutant displays additional divisions in the columella cell lineage, promoting root cap disorganization, formation of additional cell layers below the QC, and reduced emergence of lateral roots in Arabidopsis (De Smet et al., 2008). Hence, ACR4 is critical for completing the RAM appearance phase, confirming its role in the maintenance of QC integrity.

Primary root caps exhibit a cuticle only very early on, suggesting that the definition of root developmental and physiological traits occurs during this initial period. Later, this structure was found exclusively in the root caps of lateral roots at the start of the root-foraging phase in Arabidopsis (Berhin et al., 2019). Taken together, these results indicate that the RAM appearance phase is characterized by elevated cell division rates and strong QC identity. It seems reasonable to assume that reduced nutrient uptake per unit root length is linked to the absence of branches, which increase the root surface area and harbor nutrient transporters. Later on, roots experience an exponential increase in nutrient uptake, which most likely arises from improved root branching status correlated with higher expression of nutrient transporter genes and complete differentiation of xylem and phloem, facilitating nutrient transport across the whole plant.

**Root-foraging phase**

Transition to the root-foraging phase follows progressive patterns of cell differentiation. During this phase, the root structure begins to become differentiated, displaying segments beyond the seminal root. Roots may emit protrusions along a longitudinal plane, which often coincides with the emergence of lateral roots, while in other cases, adventitious roots may appear first (Figure 1). At this point, the cell layers of roots may display increasing variability and greater complexity than those of shoots. Minimum requirements are likely to be necessary to induce root branching and promote cell differentiation in specific root zones. ROOT HAIR DEFECTIVE 2 (RHD2) mediates Ca^{2+} influx into the cytoplasm in response to reactive oxygen species and initiates root branching in Arabidopsis (Takeda et al., 2008). To sustain this event, SCR and SHR induce cell differentiation in roots, generating a mature endodermis with a distinguishable Casparian strip in Arabidopsis (Drapek et al., 2017). Intriguingly, feed-forward loops point to a transcription factor activation cascade, whereby SCR and SHR bound to regulatory regions induce the expression of a third transcription factor, MYB.
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DOMINANT PROTEIN 36 (MYB36). This event results in the transition from proliferation to differentiation of endodermal cells, culminating in the formation of the Casparian strip in Arabidopsis (Liberman et al., 2015). In this process, SHORT HYPOCOTYL 2 (SHY2) plays a role in the repression of auxin signaling by reducing PIN-FORMED (PIN) transporter activity, which further stimulates endodermal cell differentiation in Arabidopsis (Dello Ioio et al., 2008; Di Mambro et al., 2017). Accordingly, in combination with stele-derived small peptides, SHR serves as a hub factor regulating the signaling cascade that reconstitutes Casparian strips using non-endodermal cells and employing both apoplastic and symplastic communication to coordinate root cell fates in Arabidopsis (Li et al., 2019). CIF2 promotes SHR stabilization, which, together with MYB36, promotes the acquisition of endodermis identity. In addition, roots treated with CIF2 showed increased root hair density in Arabidopsis (Drapek et al., 2018), suggesting a role for this peptide in switching to the foraging phase.

Passage cells are isolated unsuberized cells associated with meristematic patterning that exist in most angiosperms. Furthermore, older roots exhibit passage cells in the endodermis, which is characterized by thin walls and Casparian strips in Arabidopsis (Andersen et al., 2018). They play a prominent role in differential auxin and cytokinins responses inside the late endodermis circumference. Cytokinin inhibition triggers almost complete loss of suberization, which correlates with endodermal cells becoming passage cells. Nevertheless, suberization probably persists around lateral root emergence sites, indicating insensitivity to cytokinins in Arabidopsis (Andersen et al., 2018). Reflecting a dynamic induction of the foraging phase, the transcription factor ROOT HAIR DEFECTIVE SIX-LIKE4 (RSL4) is synthesized in a 4-h pulse during root hair cell elongation and is then gradually degraded via the 26S proteasome in Arabidopsis (Datta et al., 2015). RSL4 synthesis is enhanced in response to low nutrient availability, resulting in a prolonged growth phase and the formation of long root hairs (Datta et al., 2019). The genetic and biochemical bases of root branching remain much less understood than shoot vegetative changes. Despite the existence of a root clock that positions the lateral root primordia in Arabidopsis (Marin et al., 2010; Wachsman et al., 2020; Xuan et al., 2020; Perianez-Rodriguez et al., 2021), the mechanism that leads from branching to root secondary growth requires further investigation.

Root-senescence phase

In the last phase, senescence may be progressively observed across the entire root. Cell proliferation and differentiation decline, while cell death rates increase, ultimately culminating in plant death (Figure 1). Compared with leaves and petals, little is known about senescence in roots, even though morphological and cytological changes have been documented (Wojciechowska et al., 2018). It is important to mention that perennial species may live for decades, centuries, or even millennia and, therefore, may experience the yearly renewal and selective removal of root structures. As there seems to be little association between leaf and root turnover timing in biennial or perennial species (Heilmeier et al., 1986; Withington et al., 2006; Thomas, 2013), it is possible that root senescence is regulated independently from the shoot senescence program. This would imply the occurrence of unique developmental phases in roots in a species-specific manner.

The indeterminate meristem growth found in perennial species raises doubts about whether senescence occurs at the whole-plant level, even though cellular and leaf senescence may occur simultaneously (Munne-Bosch, 2008). Trees are usually characterized by the seasonal induction of root senescence and then shoot senescence, with the same pattern also applied to suppression of senescence (Munne-Bosch, 2007). In roots, the balance between growth and senescence is likely to be a pivotal trait that defines tree mortality in temperate forests (Munne-Bosch, 2014). By evaluating the root turnover strategies of 19 monocot species, Courchesne et al. (2020) observed that species characterized by root overwintering blocked the development of new lateral roots during the late growing season. This strategy allowed them to preserve more nutrients and carbon than species in which root senescence occurred in the autumn. Likewise, herbaceous perennial species with senescent leaves manifest different root-senescence strategies. In Sparganium emersum, root senescence follows leaf senescence, whereas Iris versicolor does not display any root-senescence symptoms despite showing leaf senescence over winter (Ryser et al., 2020). Moreover, roots from perennial plants are usually characterized by color shifts, as in Vitis labrusca and Populus trichocarpa, that are likely to be associated with lower cell viability (Comas et al., 2000; Bagniewska-Zadworna et al., 2014). P. trichocarpa exhibits seasonal senescence of fine roots, akin to the vascular cell death of leaves and petals (Bagniewska-Zadworna et al., 2014; Wojciechowska et al., 2018). In the perennial species Trifolium pratense, a decline in longevity and respiration, as well as a loss of membrane integrity in roots, correlates positively with the loss of cell viability and negatively with a lower sugar content (Bingham and Rees, 2008; Webb et al., 2010; Bingham, 2012). In perennial species, N levels determine the overall content of live and dead fine roots (Kunkle et al., 2009). Notably, both the size of the meristem and its activity are highly relevant for senescence in shrubs and trees (Munne-Bosch, 2007). Therefore, root development seems to be involved in the senescence regulation of perennials, and organ size is relevant for the acquisition of senescence competence in annual plants.

Annual and biennial plant species must develop the competence to initiate senescence, with a terminal growth phase characterized by declining physiological efficiency. Ethylene becomes less effective in inducing senescence in older plants (Jing et al., 2005), indicating that once the plant acquires the competence to senesce, acceleration of this process by exogenous factors is rather complex. Senescence has been demonstrated to progress differently across root zones in annual species (Tsimilisina et al., 2019). QC cells are central modulators of senescence, as their size becomes smaller and variations in cell divisions occur with root age (Tsimilisina et al., 2019), indicating a constant decline in QC identity (Figure 1). It has been recently demonstrated that root senescence may be explained by a gradual decrease in WUSCHEL-RELATED HOMEBOX 5 (WOX5) expression over time and consequent decline in QC identity in Arabidopsis (Wein et al., 2020). Because diverse structures and zones of maize roots display differences in natural senescence, the latter is likely to be induced by aging and not in
response to environmental conditions. The maize root cortex is characterized by abundant cell viability over the plant life cycle, with root hairs exhibiting a lifespan of 2–3 days and constant renewal until the late grain-filling stage (Fusseder, 1987). This suggests that the entire root system enters the terminal senescence stage. Cell death in the cortex and stele of axial maize roots follows the onset of shoot senescence, exhibiting strict developmental synchrony (Wenzel and McCully, 1991). Liu et al. (2019a) reported root-specific senescence-related transcription factors in barley, some of which were not expressed in leaves. This finding suggests that roots undergo an intrinsic, genetically determined senescence program that is influenced by aging.

Cell death occurs throughout root development, during which root ontogeny regulation exhibits a noticeable dependence on PCD, which promotes active cell elimination (Bagniewska-Zadworna and Arasimowicz-Jelonek, 2016). Although ontogeny plays an important role in the regulation of root transcriptional gradients in Arabidopsis (Wendrich et al., 2017), its effect on PCD and senescence in distinct root zones has been less well explored. Senescent organs are defined by limited carbon availability, which is the case for roots during aging (Kosola et al., 2002). The shoot-derived carbon pool in roots has direct implications for root aging and turnover of Sassafras albicum (Adams and Eissenstat, 2014), and its remobilization is important during senescence. Intriguingly, PCD-dependent root cortical senescence in Triticaceae species acts antagonistically to ethylene and lowers root respiration rates (Schneider et al., 2017a, 2017b, 2018). This finding highlights the phenomena that may follow PCD during senescence. Whereas elevated cell-death rates in the primary root tip trigger auxin release into proximal tissues, thereby determining root cap turnover and root foraging in Arabidopsis (Xuan et al., 2016), it remains unclear whether lateral roots exhibit senescence patterns similar to those of leaves in the shoot.

Similar to shoot developmental phases, the final stage in the root life cycle also seems to be associated with a loss of RAM quiescence. Specifically, the reduced ability of cells in the RAM QC to proliferate probably promotes the terminal growth of roots. Overall, remarkable physiological changes seem to occur during root senescence, including limitations in carbon availability and respiration, remobilization of compounds, and increasing rates of cell death, in which ethylene is a pivotal triggering factor. Hence, a hormonal network seems to modulate root senescence as well as its early development and phase progression, but it is not clear how senescence is triggered and whether or not it is determined.

ENDOGENOUS AND EXOGENOUS CELL CUES REWIRE ROOT PHASES

Distinct developmental pathways fine-tune cell fate in response to endogenous and environmental cues. During development, this mostly involves cell-cycle regulation to ensure appropriate cell proliferation and differentiation rates. A comparison of cell-cycle duration (T) in 17 species revealed very narrow variations in T among roots from the same species, irrespective of age or growth conditions (Zhukovskaya et al., 2018). Root systems of plants that originated from the same genotype exhibited strong developmental overlap with those of other rice genotypes (Fang et al., 2013). By contrast, root systems that belong to distinct genotypes tend to avoid one another while growing in the same soil region (Fang et al., 2013). Changes in cell size around the root meristem denote the onset of cell differentiation and identity acquisition, whereby root tips sense contrasting communication inputs to modulate root architecture in rice and Arabidopsis (Fang et al., 2013; Pacifici et al., 2018). Application of the synthetic clavata3/ESR-related 40 (CLE40) peptide causes meristem cell differentiation in Arabidopsis, rice, and barley, whereas columella stem cells become differentiated only in Arabidopsis (Kirschner et al., 2017). Thus, divergent pathways seem to support the maintenance of differentiation status in distal stem cell populations, indicating the existence of unrelated developmental pathways in the roots of different species.

Time and space can provide appropriate guidance for plant organ growth and architecture, forming intrinsic relationships with the environment to determine cell fates around meristematic zones (Scheres and van der Putten, 2017). Key mediators of root development, such as RETINOBLASTOMA-RELATED 1 (RBR1), WOX5, NAC PROTEIN FEZ, SOMBERO, AUXIN RESPONSE FACTOR 10 (ARF10), and ARF16, provide independent inputs to regulate root development in Arabidopsis (Bennett et al., 2014). The autonomous, gibberellin, photoperiod, and temperature (vernalization) pathways act independently during developmental phase transitions in shoots, particularly during flowering. Interestingly, some of the cues that regulate shoot development behave similarly during phase progression in roots.

Cell-autonomous pathway

RBR1 plays a crucial role in the maintenance of stem cell integrity and performs unique functions in different cell types around the meristem of Arabidopsis (Wachsmann et al., 2011). Specifically, RBR1 activity limits the division of columella cells and favors their differentiation, revealing cell-autonomous behavior in distinct cell types (Wachsmann et al., 2011). SCR acts in a cell-autonomous manner, regulating fates around QC cells, where it interacts with RBR1 to module de novo cell formation in Arabidopsis (Sabatini et al., 2003; Cruz-Ramirez et al., 2013). Similarly, both MYB36 mRNA and protein display autonomous patterns in a small group of cells surrounding the lateral root primordia and participate in the transition toward the root-foraging phase in Arabidopsis (Fernández-Marcos et al., 2017). DNA methylation of specific root cell types, particularly those in the columella root cap, was shown to mediate root development through heterochromatin loss in Arabidopsis (Kawakatsu et al., 2016). As the expression gradient of PLETHORA (PLT) transcription factors declines in line with HISTONE ACETYLTRANSFERASE GCN5 activity in the columella stem cell layers, differentiation of the corresponding cells may be attenuated by differential histone acetylation in Arabidopsis (Kornet and Scheres, 2008). Local gene expression and protein activity indicate the existence of an autonomous pathway that promotes the induction of an early foraging phase (Figure 2). By contrast, mobile elements (e.g., mRNA, proteins, and hormones) may participate in the non-autonomous regulation of developmental pathways.

Non-cell-autonomous pathway

Certain elements can move across different root cell types in Arabidopsis, rice, and Brachypodium distachyon (Wu et al.,...
suggesting the existence of a developmental pathway based on non-cell-autonomous regulation (Figure 2). For example, SHR is limited to the endodermis owing to inhibition by SCR, a conserved mechanism that favors a single endodermis in roots of Arabidopsis (Cui et al., 2007). By contrast, JACKDAW (JKD) is localized around QC cells, where it regulates asymmetric cell divisions and SHR movement, as well as the gene expression of the epidermal cell-fate regulators GLABRA 2, CAPRICE, and WEREWOLF in Arabidopsis (Hassan et al., 2010). Non-cell-autonomous patterns revealed by JKD suggest that this protein affects signaling throughout the cortex, particularly in root hair cells (Hassan et al., 2010). Both miR165a and miR166b exhibit mobility in the roots similar to that in the SAM, promoting the degradation of mRNAs encoding transcription factors that regulate endodermis and stele cell fates in Arabidopsis (Carlsbecker et al., 2010). When SHR moves into the endodermis and activates SCR, it enables miR165/6 to move and complete the non-cell-autonomous network (Carlsbecker et al., 2010). This suppresses PHABULOSA (PHB) gene expression in the

Figure 2. Pathways that promote root developmental phase transitions in Arabidopsis thaliana.

(A) The autonomous pathway appears to promote the root-foraging phase, during which the transcription factor MYB36 mRNA is found only in specific cell types, whereas the MYB36 protein induces genes to promote foraging. Similarly, PLETHORA (PLT) expression varies across root zones and is highest around root stem cells, where differential DNA methylation patterns may promote maximum PLT activity. In these cells, RETINOBlastoma-related 1 (RBR1) and SCARECROW (SCR) regulate cell fates around QC cells, promoting de novo formative cell divisions.

(B) Mobile elements that move among diverse root cell types constitute the non-autonomous pathway. SHORT-ROOT (SHR) moves from the epidermis to the endodermis, where SHR activates SCR to induce the movement of miR165/6, which in turn suppresses the expression of cell-differentiation regulators to release the root-foraging phase. JACKDAW (JKD) activity around QC cells delimits SHR movement in these cells, whereas the expression of genes encoding GLABRA2 (GL2), CAPRICE (CPC), and WEREWOLF (WER) is regulated by JKD, resulting in the mobility of these three genes to give rise to the epidermis. Overall, the non-autonomous pathway sustains the root-foraging phase (from the middle of the phase), vascular tissue formation, and growth.

(C) Hormones are essential to organism development, and an emerging hormonal pathway orchestrates root developmental phases. Thus, opposing gradients of auxin and cytokinin regulate patterns of cell proliferation and differentiation. In the root tip, maximum levels of auxin with jasmonates may induce the transcription factor ETHYLENE RESPONSE FACTOR 109 (ERF109) that in turn activates QC cells to promote stem cell fates, particularly around the root tip. Subsequently, a local auxin maximum at the pericycle cells activates CLASS III HOMEODOMAIN-LEUCINE ZIPPER (HD-ZIP III) transcription factors, and increases in auxin and HD-ZIP IIs lead to the acquisition of xylem identity. As a consequence, ERF109 activity is upregulated at the transition zone according to the balance of jasmonate and auxin, thereby inducing lateral root (LR) formation. Therefore, the hormonal pathway regulates not only the early and later stages of the foraging phase but also entry into the senescence phase.

(D) Roots contain cells that vary markedly in ontogeny and require complex genetic circuits to regulate developmental phases, suggesting a periodic pathway. The condensation of AUXIN RESPONSE FACTOR (ARF) transcription factors in the cytoplasm, where they are inactivated, limits auxin responsiveness. Similarly, cyclic cell death of epidermal cells in the transition zone results in periodic LR induction. Meanwhile, the GNOM protein, a vesicle trafficking regulator, is repressed by ARF-GTP ACTIVATING PROTEIN DOMAIN (AGD3), and both proteins regulate the balance between esterification and de-esterification, orienting the turnover of pectin esterification. In the oscillation zone, a root clock is established through a competitive interaction between GNOM and AGD3 that orients the functionality of the LR clock to promote the root-foraging phase. Taken together, evidence suggests that the periodic pathway regulates the overall phases of root development from early until late cell development.
Exploring root developmental phases

In plants, signal transduction inputs follow distinct hormonal leads through tissues, enabling the sequential development of various structures. Regulation of stem cell fate in roots (Figure 2) relies on a tight balance between independent hormonal inputs that promote root development in Arabidopsis (Bennett et al., 2014). A dynamic hormonal network in multiple cell types around the RAM regulates cell fate and tissue patterning. Specifically, auxin may concurrently induce and inhibit WOX5 expression in distinct cell types of Arabidopsis (García-Gómez et al., 2017). In the embryonic stage, auxin controls cytokinin activation at specific sites by promoting cell divisions that allow vascular tissue growth and by organizing layer patterns in Arabidopsis (De Rybel et al., 2014b). The initial steps in Arabidopsis root cell differentiation require changes in cell size; hence, cytokinin indirectly modulates cell-wall remodeling to drive cell differentiation (Pacifici et al., 2018). Cytokinin oxidase 2 (CKX2) controls cytokinin degradation and regulates the root system’s radial distribution patterns based on anti-gravitropic signaling in Arabidopsis (Waidmann et al., 2019). The root transition zone separates dividing and differentiating cells, and the hormone cytokinin regulates auxin polar transport and degradation (Di Mambro et al., 2017). The ensuing basal auxin profile establishes the transition zone, defining division and differentiation layers in Arabidopsis (Di Mambro et al., 2017). A local auxin maximum acts with CLASS III HOMEODOMAIN-LEUCINE ZIPPER (HD-ZIP III) transcription factors to regulate xylem identity acquisition and form a stem cell organizer with cellular quiescence in Arabidopsis (Smetana et al., 2019). Thus, auxin seems to be particularly important for the root-forming phase. In addition, its dynamic behavior modulates Arabidopsis root elongation in the different root zones following repression of cytokinin signaling (Andersen et al., 2018). ABA repression culminates in the absence of suberin in passage cell precursors (Andersen et al., 2018), demonstrating how hormonal balance enforces differential meristem patterning regulation. Following tissue formation and patterning establishment, root cells have to cope with a large number of damaging agents that impair growth and development and may be overcome by regeneration pathways. In Arabidopsis roots, cell replacement is guided by dynamic auxin and cytokinin domains that follow the embryonic stage (Efroni et al., 2016). By contrast, the formation of a new ground tissue layer specifying the Arabidopsis middle cortex is precisely regulated by gibberellin and SCR via an additive effect and, independently, by SHR (Paquette and Benfey, 2005). Intriguingly, restorative divisions after damage and middle cortex formation do not seem to respond to gibberellin, suggesting that a different mechanism mediates these processes in Arabidopsis (Marhava et al., 2019). ETHYLENE RESPONSE FACTOR 109 (ERF109) acts at the interface between jasmonate signaling and auxin biosynthesis and transport, mediating Arabidopsis lateral root formation (Cai et al., 2014; Liu et al., 2019b) and subsequent foraging. In addition, ERF109, ERF115, and jasmonate regulate cell-division proteins to orient the stem cell niche fates of Arabidopsis (Zhou et al., 2019).

In summary, multiple elaborate hormonal networks mediate distinct transition steps, indicating that hormones have a multifaceted role in root development, coordinating not only early and later stages of the foraging phase but also entry into the senescence phase (Figure 2).

Periodic pathway

Oscillations in the expression and activity of genes, proteins, and metabolites promote tissue patterning in roots, suggesting that periodic events regulate the development of root structures. Root cap cells release auxin periodically over time, controlling a switch between developmental phases in these cells (Laskowski and Ten Tusscher, 2017; Möller et al., 2017). The different root zones exhibit contrasting developmental times, hinting at periodic genetic and physiological regulation (Figure 2). Cell shape is determined through local positive feedback in root hair cells, with Ca²⁺ and RHD2 interacting at the growing point of the root section in Arabidopsis (Takeda et al., 2008). Therefore, growth location and stability at specific sites anticipate cell fate (Takeda et al., 2008). By contrast, the aggregation of ARF transcription factors causes cytoplasmic localization and inactivation, limiting auxin responsiveness in distinct root zones in Arabidopsis (Powers et al., 2019). Thus, the lateral root cap displays a multilayered structure that does not obey the correspondence between cell age and distance from the QC. This contrasts with other root tissues and violates the spatiotemporal endoploidy model of the lateral root cap in Arabidopsis (Bhosale et al., 2018a). Cyclic transcriptional pulses establish periodic developmental regulation, which contributes to root pre-branch formation in Arabidopsis (Moreno-Risueno et al., 2010). Similarly, cyclic PCD enhances hormonal signaling, resulting in periodic lateral root induction and optimization of water and nutrient uptake in Arabidopsis (Xuan et al., 2016).

Multiple tissue-forming steps are regulated by the same set of transcription factors and their positional cues (Moreno-Risueno et al., 2010). Therefore, competition for the gradual transcriptional regulation of cell differentiation in Arabidopsis (Wendrich et al., 2017) seems to depend on cell ontogeny. Periodic pre-branch formation exhibits a lateral root clock that follows a specific spatial pattern in Arabidopsis (Van Norman et al., 2014). The vesicle trafficking regulator GNOM and the
INVolVEMENT OF DEVELOPMENTAL PHASEs IN ROOT PLASTICITY

Plants experience extremely contrasting environmental conditions. Whereas shoots exchange most of the gases, roots exchange most of the remaining compounds because of their more aqueous external environment. The fossil roots of the lycopsid *Asterozyxylon mackiei* revealed an absence of root caps; instead, a continuous epidermis covered the entire meristem surface (Hetherington and Dolan, 2018). Hence, root cap acquisition is defined as a crucial moment in evolution that facilitated plant terrestrialization. Another feature that explains plant growth under oscillatory or extreme environments is root endoreplication across diverse cell types, which alters nuclear and cell-wall features. Coordinated cell expansion highlights the contrasting characteristics of lateral root caps compared with most other root tissues of *Arabidopsis* (Bhosale et al., 2018a). Shifts in cell structure are coupled with cell division and expansion, enabling developmental progression.

The primary root cap cuticle promotes meristem protection during germination and facilitates the emergence and invasive growth of lateral roots during foraging in *Arabidopsis* (Berhin et al., 2019). The suberization layer is deposited in response to a range of nutrients by directing functional and anatomical plasticity in the adult endodermis of *Arabidopsis* (Barberon et al., 2016). Although the number of cells in this layer tends to remain constant, suberization changes may occur in accordance with hormonal (ethylene and ABA) patterns. Thus, diffusion barriers in the endodermis respond to Casparian strip plasticity and differential suberization patterns (Figure 3A). This promotes delays in apoplastic barrier establishment and the arrest of cell development in *CASPARIAN STRIP MEMBRANE DOMAIN PROTEIN* (CASP) mutants and particularly in *EXOCYST COMPLEX* (EXO) *Arabidopsis* mutants (Kalmbach et al., 2017). Recently, a specific gene encoding an exocytosis factor corresponding to the EXO 70 subunit was identified in *Arabidopsis*. This gene modulates root system architecture and depth and affects root tip auxin transport, orienting root gravitropism to cope with variable rainfall patterns (Ogura et al., 2019). The origin and evolution of rapid root gravitropism were attributed to changes in amyloplast patterns within the root apex (Figure 3B). These patterns promote variations in the auxin transporter PIN2, whose alteration of auxin flow dynamics links gravity perception in the root cap to cell expansion in the root elongation zone in *Arabidopsis* (Zhang et al., 2019b). These molecular players promote and regulate root plasticity to improve the uptake of water and nutrients through precise communication between the root and rhizosphere.

Root tips can recognize soil obstacles by two means: (1) root-root recognition following root exudate patterns; and (2) root-object recognition through physical contact. Crucially, both alter root architecture (Fang et al., 2013) and guide the response to soil texture, as exemplified by rice genotypes that respond with differential plasticity to soil particle variations (Rogers et al., 2016). Root foraging is modulated by water availability in distinct root zones to prevent the emergence of lateral roots at sites with low water availability (Figure 3C). This hydropatterning strategy depends on water potential gradients across the root and results in differential lateral root growth (Robbins and Dinneny, 2018). Hydropatterning alters root architecture in response to water contact, eliciting SUMOylation of the auxin response factor ARF7 on air-exposed root sides of *Arabidopsis* (Orosa-Puente et al., 2018). This post-translational modification promotes the interaction between ARF7 and the auxin-responsive protein IAA3, which represses LATERAL ORGAN BOUNDARIES DOMAIN 16 (LBD16) and prevents root branching initiation (Orosa-Puente et al., 2018). At the same time, lateral root formation is repressed and cell-identity acquisition is blocked by ABA in soil air pockets under water scarcity in *Arabidopsis*, barley, and maize (Orman-Ligeza et al., 2018). Thus, the root-foraging phase seems to react simultaneously to environmental cues via hydropatterning and to endogenous pathways via an on/off or rewiring response (Figure 3C). In this sense, root plasticity has a remarkable potential to trigger the regeneration of cells and tissues.

A complex conceptual framework defines regeneration and structural changes. To regenerate roots after complete root stem cell excision, complementary hormonal domains are established in root zones, forming sites that recapitulate embryogenesis steps and culminate in de novo generation of stem cells (Efroni et al., 2016). This ability allows the root primordium to regenerate the shoot meristem, with shoot removal triggering the root greening response that might otherwise be repressed by auxin signals in *Arabidopsis* (Kobayashi et al., 2017). According to this new concept, root cell identity transitions rely on positional and/or hormonal cues to identify differentiation patterns (Sugimoto et al., 2011; Efroni et al., 2016).

Lateral roots are sites of ongoing cell differentiation. WOX11 regulates root plasticity and mediates rooting in leaf and stem explants as well as hypocotyls. This, in turn, promotes proper rooting upon wounding and drought in *Arabidopsis* (Sheng et al., 2017). Accordingly, PLT transcription factors promote cell division to regulate de novo meristem formation and coordinate tissue patterning during lateral root outgrowth of *Arabidopsis*.
Exploring root developmental phases

Figure 3. Root developmental phases and plasticity.

(A) Roots growing under natural conditions may experience salt stress. Under these conditions, abscisic acid (ABA) triggers suberization of the endodermis, thereby blocking the entry and exit of substances.

(B) Roots exhibit a protective structure composed of cutin polyesters (orange caps) that protects the meristem during early development; this structure also protects lateral roots during the foraging phase. Gravitropism has a remarkable effect on EXOCYST70A3 protein activity that in turn modulates the PIN3 transporter, which reorients auxin flux to shift the original elongation plane of the root. Similarly, gravity mediates changes in the amyloplast patterns of root apex cell-group populations, altering the orientation of auxin flow in accordance with PIN2 transporter activity and thereby modifying the root elongation plane.

(C) Variations in water potential ($J_w$) induce distinct developmental patterns in roots. Higher $J_w$ promotes optimal activity of AUXIN RESPONSE FACTOR 7 (ARF7), inducing the LATERAL ORGAN BOUNDARIES DOMAIN 16 (LBD16) gene and promoting the root-foraging phase. By contrast, regions of the rhizosphere that are rich in air spaces enhance the SUMOylation of ARF7 (SUMO), thereby recruiting IAA3, repressing LBD16 expression, and arresting root foraging. In these spaces, ABA accumulation impairs cell-identity acquisition, blocking pre-branch site formation.

(D) Cell ablation and tissue damage are repaired by means of particular events. Convergent gradients of PLETHORA (PLT) transcription factors and cell regeneration competency are observed over the root regions, and maximum levels of both are found around QC cells. Under optimal growth conditions, PLT may regulate formative cell-proliferation patterns (the common cell cycle with four phases: G1, synthesis [S], G2, and mitosis [M]), whereas re-activation of stem cell status requires the re-establishment of the PLT expression gradient (restoration pattern) converging to a gradient of cell regeneration competency at the damaged site. Thus, the restoration pattern shows a divergent cell-division plane for proliferation, in which cell-cycle progression is accelerated by the activation of SHORT-ROOT (SHR), SCARECROW (SCR), and CYCLIN D6;1 (CYCD6;1). This acceleration reflects a shorter duration of the cell-cycle phases (G1, S, G2, and M). Consequently, tissue damage (purple circles) activates a hormonal circuit to orchestrate tissue repair. Jasmonates may directly induce ERF115, which regulates stem cell activation and tissue regeneration, and/or induce ETHYLENE RESPONSE FACTOR (ERF) 109 and ERF115, which promote the last steps of tissue regeneration. At the same time, local auxin accumulation at damaged sites promotes the expression of ERF115, which orients stem cell activation and, ultimately, tissue regeneration.
of stem cells after cell death in Arabidopsis (Heyman et al., 2016). Hormone shifts cooperate in this process based on the type of damage (Figure 3D). Jasmonate coordinates wound signaling to enable de novo root regeneration from detached leaves via ERF109 activation and auxin biosynthesis induction in Arabidopsis (Zhang et al., 2019c). Specifically, jasmonate induces ERF109, which in turn stimulates CYCLIN D6;1 (CYCD6;1) expression and functions upstream of ERF115 to enable root regeneration of Arabidopsis (Zhou et al., 2019) (Figure 3D). Subsequently, JASMONATE ZIM-DOMAIN (JAZ) proteins inhibit ERF109, replacing constitutive regeneration with de novo regeneration (Zhang et al., 2019c). Intriguingly, these players function together with SCR and RBR to adjust stem cell niche replacement and maintain QC quiescence of Arabidopsis (Zhou et al., 2019). During stem cell reactivation, a PLT expression gradient is formed in the roots, indicating the regenerative competence of cells, together with accelerated cell-cycle progression in Arabidopsis (Marhava et al., 2019) (Figure 3D). Cell-division reprogramming for replacement differs across cell types. In general, activation of SHR, SCR, and CYCD6;1 in the endodermis or near-endodermal cortex ensures correct cell fates in Arabidopsis (Marhava et al., 2019) (Figure 3D). As a countermeasure, cells may be selectively eliminated to maximize energy and nutrient use, favoring developmental phase transitions.

Early cell descendants from root stem cells are preferentially eliminated under chilling stress. By contrast, columella stem cell daughters are highly exposed to DNA damage, and even if the directional auxin flux from these cells is disrupted, the quiescent state of the root stem cells is preserved in Arabidopsis (Hong et al., 2017). The sacrifice of these cells to sustain root survival explains how and to what extent alternative mechanisms regulate root development and direct responses to environmental cues. Thus, the precise timing of cell death is a critical step that enables cell renewal and the entry of cells into the root elongation zone to ensure root growth and homeostasis in Arabidopsis (Fendrych et al., 2014). In short, plasticity allows the roots to cope with soil dynamics through hormone-mediated changes in root structure, architecture, and regeneration.

**NUTRITIONAL CHECKPOINTS GOVERNING ROOT DEVELOPMENTAL TRANSITIONS**

The most relevant environmental inputs that affect root development include water and nutrient uptake from the soil. Water is lost to the atmosphere through the stomata via several interconnected pathways, whereas nutrient uptake, transport, and use are regulated by numerous complex pathways. Agricultural revolutions over the last century have remarkably improved crop productivity, and plant breeding has focused on controlling shoot productivity, and plant breeding has focused on controlling shoot architecture and architectural characteristics can reduce nutrient imbalances (Cormier et al., 2016). The scarcity of some fertilizers as well as their elevated production costs and environmental impact have limited sustainable agriculture. Crop yield is largely dependent on sufficient N and P. Because N fertilizers volatilize easily and release N₂ into the atmosphere, large amounts are required to sustain plant growth, whereas P is immobile and highly adsorbed in the soil. Although substantial advances have been made in understanding the mechanisms of uptake, transport, and efficient use of both N and P, only improved knowledge of how root developmental phase transitions respond to nutrients will help generate crops capable of more efficient nutrient use.

**N modulates early root developmental phases**

Nucleotides, amino acids, and proteins required for plant growth and development contain N, which is estimated to consume almost 2% of global energy, and N runoff leads to the contamination of lakes and rivers (Sutton et al., 2013). In the soil, N is available mostly as ammonium (NH₄⁺) and nitrate (NO₃⁻), which stimulate contrasting pathways associated with root development (Motte et al., 2019; Meier et al., 2020). NH₄⁺ blocks primary root growth, decreasing both meristem length and the number of proliferating cells, as well as reducing the root elongation zone in Arabidopsis (Liu et al., 2013). Successful seed germination depends on NO₃⁻ via NIN-like protein 8 (NLP8) and ABA catabolism in Arabidopsis (Yan et al., 2016). Similarly, the transcription factor teosinte branched 1/cycloidea/proliferating cell factor 1-20 (TCP20), which is implicated in Arabidopsis embryo development, responds to NO₃⁻ levels by preferentially impairing lateral root growth (Guan et al., 2014) and the root-foraging phase. Arabidopsis TCP20 interacts with both PLTs and SCR to precisely regulate WOX5 transcripts (Shimotohno et al., 2018), indicating that NO₃⁻ regulates the root transition from the embryonic to the RAM appearance phase (Figure 4). CLE peptides and CLAVATA1 (CLV1) receptors are fundamental for triggering root branching in response to N levels in the rhizosphere of Arabidopsis (Araya et al., 2014). CLE1, CLE3, and CLE7 are expressed preferentially in pericycle cells, whereas CLV1 is present in phloem companion cells (Figure 4). Under NO₃⁻ deficit, these cells increase the expression of CLE peptides and CLV1 transcripts to halt root branching (Araya et al., 2014). Furthermore, auxin derived from shoots accumulates in root regions previously supplemented with NH₄⁺, inducing the emergence of lateral roots in Arabidopsis to mediate root foraging along the “soil” surface (Meier et al., 2020).

A combinatorial network of interactions between N forms and hormones (auxin, cytokinin, and ABA) has been described in Arabidopsis roots, revealing the exact transcriptional regulation that drives ABA and NO₃⁻ responses (Ristova et al., 2016). Another study highlighted the role of cytokinin signaling in Arabidopsis responses to N demand (Ruffel et al., 2011). In brief, cytokinins induce GLUTAREDOXIN (GRX) expression to repress primary root elongation under elevated soil NO₃⁻ levels while at the same time promoting root branching in Arabidopsis (Patterson et al., 2016). The localized exposure of specific root sections to high NO₃⁻ levels triggers genes involved in both cell-cycle progression and auxin efflux from the phloem to pericycle cells, further promoting root branching in maize (Yu et al., 2015b). Root-specific cell types experience particular transcriptional rearrangements based on differential NO₃⁻ content along the root length (Yu et al., 2015b). Thus, differential root branching patterns in roots allow plants to explore heterogeneous NO₃⁻ environments. Reduced root branching under low N conditions may result in greater N capture in maize (Zhan and Lynch, 2015),
suggesting that a tailored root developmental program might improve N capture and utilization with reduced energy expenditure.

Soils are highly dynamic environments that impose diverse interactions among nutrients and physical, chemical, and biological components. A non-uniform root distribution pattern in the soil enables precise N sensing. Moreover, small C-terminally encoded peptides (CEPs) mediate long-distance communication, linking roots and shoots of *Arabidopsis* under N starvation conditions (Tabata et al., 2014). CEP1 and CEP5 are expressed mostly in the basal region of lateral roots, where elevated levels of these genes repress primary root growth (Tabata et al., 2014). Overall, multiple strategies to rewire the developmental program allow plants to cope with N starvation and other nutritional disturbances. Several avenues must be explored to better understand root development in response to N fluctuations. Considerable engineering feats will determine the precise transcriptional and post-translational regulation of multiple genes that may shape root system architecture differentially across soil gradients.

**On the connections between P and late developmental phase transitions of roots**

Intensive food production over the past centuries has significantly altered the structure and chemical nature of soils and has modified their retention capacity for certain nutrients. In particular, P sources have declined drastically, endangering food security. Most arable soils in the world have an acidic pH, under which P is in stable and difficult-to-assimilate forms: dihydrogen phosphate (H$_2$PO$_4^-$) and hydrogen phosphate (HPO$_4^{2-}$) (Kochian et al., 2004; López-Arredondo et al., 2014). Application of fertilizers may not be sufficient to meet the demand for P in crops. Hence, enormous efforts are being made to improve the efficiency of P uptake and its use by plants. Understanding how developmental root phases adjust in response to P is likely to be fundamental to ensuring food security.

Passage cells are commonly maintained in an unsuberized state, but this may change in response to P during meristem patterning, promoting expression of the P efflux protein PHO1 (Andersen et al., 2018). Consequently, vascular cell proliferation in roots is
coordinated by the gene target of MONOPTEROS 5 AND LONESOME HIGHWAY (TMOS/LHW), which promotes the biosynthesis of mobile cytokinins in Arabidopsis (Andersen et al., 2018; Wendrich et al., 2020). TMOS/LHW protein re- orients both the length and the fate of epidermal cells to increase branching under low P conditions, indicating the important role of this protein in the response to P limitation during foraging. Under low phosphate conditions, synthesis of the transcription factor RSL4 surpasses its degradation, causing the formation of long root hairs and demonstrating how protein stability determines hair size in Arabidopsis (Datta et al., 2019). In this context, auxin transport from the root apex to the differentiation zone in response to low P promotes root hair development, which is associated with the induction of the transcription factors ARF19, RSL2, and RSL4 in Arabidopsis (Bhosale et al., 2018b). Under low external P, the root auxin influx transporter OsAUX1 alters the root angle and promotes preferential foraging in the top soil layers, although this is not sufficient to improve P uptake (Giri et al., 2018). AUX1 is the main transporter for auxin in root hairs, facilitating membrane depolarization at acidic extracellular pH and sensing external P in Arabidopsis (Dindas et al., 2018). Moreover, P limitation in the soil reduces secondary root growth while enhancing P uptake, a phenomenon that varies according to the genetic background in common bean (Phaseolus vulgaris) (Strock et al., 2018). P is likely to be a key factor in the transition from RAM appearance to the root-foraging phase (Figure 4).

Taking into account the complexity of soil and the impact of interactions among nutrients on antagonistic or synergistic outcomes regarding nutrient use, a better understanding of how nutrient dynamics modulate phase transitions in roots is needed. Rice tightly coordinates N and P utilization through interaction between the NO₃⁻ sensor NRT1.1B and the phosphate signaling repressor SPX4 (Hu et al., 2019). Arabidopsis NIGT1 is a transcriptional repressor of the NO₃⁻ transporter NRT2.1 and is itself stimulated by PHR1, the master regulator of P starvation responses, which limits nitrate uptake (Maeda et al., 2018). In maize, P and N drive root development to produce optimal lateral root branching density (Postma et al., 2014), enabling both soil foraging and root developmental plasticity. P and N modulate root elongation and branching; however, the effects of nutritional checkpoints on subsequent root developmental stages remain poorly understood. Barley root cortical senescence is ameliorated under P and N deficiency, whereby reduced respiration and nutrient content allow greater root growth (Schneider et al., 2017a, 2017b). Conserved pathways related to local and long-distance N signaling control P starvation responses in O. sativa and Triticum aestivum (Medici et al., 2019). Expanding our understanding of how nutrients modulate root developmental programs may foster sustainable agriculture by decreasing the use of correctives and fertilizers while maximizing crop yield.

**CONCLUDING REMARKS**

Over at least three decades, scientists have identified several factors that regulate developmental phase transitions in plant shoots. In comparison, much less attention has been given to root developmental phase transitions. However, this may be changing, as highlighted in the present review. We have described current evidence pointing to the existence of a unique developmental program for roots, which contrasts with that in shoots because of the absence of a reproductive phase. Root-specific development is based on tightly regulated generation and elimination of root structures. In addition, the ability of roots to alter their structure in response to the environment is associated with their regeneration capacity and gradual age-dependent loss owing to decreased QC identity.

Although the excellent publications surveyed here provide valuable blueprints of the current state of the art, fully understanding developmental root phases will require additional theoretical and experimental capacities. Further progress will be related to plant hormones, as well as nutritional elements, and their function in the regulation of root growth, development, and overall plasticity. Thus, a new era of root developmental biology is expected. Importantly, new knowledge will help maximize the use of nutrients and water by crops, thus reducing the need for correctives and fertilizers. Notably, uptake and utilization of nutrients linked to cell division (e.g., N and calcium) may be improved by prolonging early developmental phases. Instead, faster induction of the foraging phase would favor uptake and utilization of nutrients associated predominantly with cell differentiation (e.g., P and iron). In summary, the mechanisms that govern root developmental phases are now emerging. Timely investments and research are likely to bring returns in the form of an improved ability to carry out rational engineering of this “black box.”

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**AUTHOR CONTRIBUTIONS**

J.A.S. and W.C.O. conceived the project. J.A.S. wrote the manuscript. W.C.O. and W.L.A. revised the manuscript. All authors contributed to reviewing and editing of the manuscript. All authors read and approved the final manuscript.

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