Review Article

Improving phosphate use efficiency in the aquatic crop watercress (Nasturtium officinale)

Lauren Hibbert1,2 and Gail Taylor1,2,*

1 School of Biological Sciences, University of Southampton, Southampton, Hampshire, SO17 1BJ, UK
2 Department of Plant Sciences, UC Davis, Davis, CA, 95616, USA
*Corresponding author. E-mail: gtaylor@ucdavis.edu

Abstract

Watercress is a nutrient-dense leafy green crop, traditionally grown in aquatic outdoor systems and increasingly seen as well-suited for indoor hydroponic systems. However, there is concern that this crop has a detrimental impact on the environment through direct phosphate additions causing environmental pollution. Phosphate-based fertilisers are supplied to enhance crop yield, but their use may contribute to eutrophication of waterways downstream of traditional watercress farms. One option is to develop a more phosphate use efficient (PUE) crop. This review identifies the key traits for this aquatic crop (the ideotype), for future selection, marker development and breeding. Traits identified as important for PUE are (i) increased root surface area through prolific root branching and adventitious root formation, (ii) aerenchyma formation and root hair growth. Functional genomic traits for improved PUE are (iii) efficacious phosphate remobilisation and scavenging strategies and (iv) the use of alternative metabolic pathways. Key genomic targets for this aquatic crop are identified as: PHT phosphate transporter genes, global transcriptional regulators such as those of the SPX family and genes involved in galactolipid and sulfolipid biosynthesis such as MGD2/3, PECP1, PSR2, PLDζ1/2 and SQD2. Breeding for enhanced PUE in watercress will be accelerated by improved molecular genetic resources such as a full reference genome sequence that is currently in development.

Introduction

Watercress – An aquatic leafy-green crop

Watercress (Nasturtium officinale R. Br.) is a semi-aquatic plant that grows in flowing shallow freshwater and is found across Europe, Asia, the Americas, the Caribbean, New Zealand and Australia (Fig. 1) [1]. Watercress is placed within the Brassicaceae family together with several other important food crops including broccoli, kale, cabbage, and mustard.

A significant amount of commercial aquatic watercress cultivation is centred in a few locations including Florida in the USA, southern Spain and Portugal, France and the south of England, with 90% of production occurring in Dorset, Hampshire and Wiltshire [2]. These chalky areas provide nutrient-rich spring water and boreholes that directly supply the watercress beds [3].

Phosphate-rich fertiliser is used to boost crop yield; however, this presents a major challenge in watercress production since it results in direct leakage of phosphate into the waterways which have high conservation value. Excess phosphate results in eutrophication of aquatic ecosystems, a process where nutrient enrichment of water sources results in excessive algal and plant growth, and subsequent disruption of ecosystem community dynamic [4]. Approximately 90% of watercress farms in the UK are on, or upstream of, a Site of Special Scientific Interest (SSSI), increasing the pressure to minimise phosphate release [5].

Phosphate and the environment

Phosphate (P) is vital for plant survival; it forms the phosphodiester bonds that link nucleotides in nucleic acids and is critical for the structure of proteins and carbohydrate polymers, for powering cells through the release of phosphate from ATP and for regulating several metabolic pathways [6, 7]. Symptoms of P deficiency are retarded growth, increased root:shoot biomass, decreased leaf area and often dark green or purple-red colouration in severely deficient plants due to anthocyanin production [8–11].

Ninety percent of the global demand for phosphorus is used for food production, however, rock phosphate is a limited resource with estimates that reserves could be exhausted in the next 50–100 years [12–14]. In addition, most of the remaining rock phosphate reserves are...
under the control of only a few countries, with Morocco and the Western Sahara holding over 70% of the total reserves, making it sensitive to political instability [15, 16]. This, combined with increasing costs of extraction and issues of eutrophication, make reducing fertiliser use an important global driver [17–19]. The high reactivity and low solubility of phosphates make them commonly the growth-limiting nutrient for plants. Accounting for fertiliser application, approximately 30% of global crop-land area exhibits soil P deficits although global P imbalances in water sources have not been investigated to any significant extent [20].

Phosphorus in the soil
Phosphates in soils are classified into inorganic P (Pi) and organic P (Po) and fractionised based on their solubility [21, 22]. Po typically comprises 30–70% of total soil P, however this is highly variable with values up to 90% reported [23–26]. Pi derived from rocks is only present in poorly soluble forms and any soluble phosphates released in the soil are rapidly immobilised into insoluble forms by interacting with adsorbing agents such as Al and Fe oxides or Ca and Mg carbonates, such that only a small amount of P in soils is accessible to plants (primarily in the form of inorganic orthophosphate compounds such as \( H_2PO_4^- \), \( HPO_4^{2-} \) and \( PO_3^{3-} \)) [13]. The availability of P is dynamic, changing over time depending on various factors such as temperature, water content and soil pH [27–29].

Phosphorus in aquatic systems and potential for eutrophication
Like soil, P in aquatic systems is also divided into different fractions based on solubility and reactivity in aquatic systems, with dissolved orthophosphate the most bioavailable [30, 31]. P in water adsorbs to oxides and tightly binds with carbonates in the same manner as when in soil. However, the P inputs to natural water systems and the interaction with P in bed sediments is altered. This creates a dynamic source of phosphorus that transfers between particulate and dissolved forms, between bed sediments and the water column, and between dead and living material (Fig. 2). In a watercress bed, the sediment is shallow gravel and thus P uptake from water likely represents the major P source. This is reflected in a study by Cumbus and Robinson (1977) who found that a greater proportion of P was absorbed by the adventitious roots of watercress (in the water), compared to basal roots (in the sediment) [32]. However, some organic detritus held within the sediment should still be considered. Phosphate dynamics in hydroponic agricultural systems such as watercress beds have not been studied, representing a knowledge gap, but P is likely uniformly distributed due to flowing water and regular maintenance of P concentrations.

Since P retention in sediments is high, P delivery into freshwater systems is largely governed by release from point sources such as sewage treatment works (STWs), leaking septic tanks, and from excess fertiliser application [33–35]. Globally, domestic sources contribute 54% to total P inputs into freshwater systems, 38% from agriculture and 8% from industry [36]. Although substantial steps towards P reduction in freshwaters have been made over the last 50 years there is still much to be done, with only 40% of European surface waters currently in good ecological status [37, 38]. Eutrophication of watercourses is also prevalent across the UK: in the most recent analysis, 55% of river water bodies in England
Figure 2. Cycling of different phosphate (P) fractions in natural aquatic environments. Readily available P (in the form of $\text{H}_2\text{PO}_4^-$ and $\text{HPO}_4^{2-}$ ions) is taken up in the sediment by basal roots and in the water column by adventitious roots. STWs = sewage treatment works.

failed to meet the revised P standards for good ecological status [38]. Eutrophication is both an economic as well as environmental issue. In the US, the economic damage of eutrophication equates to $2.2$ billion ($\sim £1.6$ billion) annually, due to losses in recreational water use, waterfront real estate, recovery of endangered species and drinking water [39].

Naturally, phosphate levels in chalk aquifers (where watercress farms are typically found) are less than $20$ μg/l, however, inputs of phosphate rapidly increase these concentrations above P targets downstream of watercress farms [2, 5]. In the river Itchen (Hampshire, UK) where several watercress farms are located, total SRP load comes predominantly from sewage treatment works (84.2%) but watercress beds can be responsible for up to 62% of the total reactive phosphate in some chalk streams, suggesting room for improvement in P management [5]. Casey and Smith (1994) found watercress beds increased mean P concentrations which may cause undesirable growth of algae and disruption of community dynamics [2].

One important strategy to tackle this problem of eutrophication is through plant breeding. By breeding watercress varieties with improved phosphorus use efficiency (PUE), the impact of watercress farming on eutrophication could be minimised. To date, no breeding for nutrient use has been conducted in watercress even though P release represents a clear issue in watercress production.

**Improving the environmental footprint of watercress through enhanced PUE**

Phosphate use efficiency (PUE) is defined as the capacity for biomass production using the P absorbed [40, 41]. Here PUE is used as a broader term that also encompasses phosphate acquisition efficiency, defined as the ability to take up P, as has been used in several studies [42–45]. Plant traits underpinning PUE can be observed at the macroscopic, microscopic, and molecular levels and we consider their relevance to future breeding for enhanced PUE.

To date, knowledge on P acquisition by aquatic plants only covers the effectiveness of plants for phytoremediation (removal of toxic contaminants by plants), rather than breeding for PUE in aquatic crops such as water chestnut (Eleocharis dulcis), water spinach (Ipomoea aquatica), lotus (Nelumbo nucifera) and watercress. Present information does not cover morphological or genetic components to improve PUE in aquatic species, and with new plant species emerging as suggested model organisms, watercress is offered as a model crop for aquatic systems [46–48]. The need for aquatic model
crops is only exacerbated by increasing market value in indoor hydroponic cultivation systems [49].

**Traits for improving P acquisition**

**Macroscopic traits: Root structural architecture (RSA)**

Root structural architecture (RSA) defines the spatial configuration of the root system and variation in RSA can reflect efficient phosphate uptake by plants [50]. Much of the current literature focuses on RSA traits in maize (*Zea mays*), common bean (*Phaseolus vulgaris*) and *Arabidopsis thaliana* with RSA shown to be a highly plastic trait, changing in response to the availability of water, nutrients and hormone signalling [51–53]. For example, ethylene is involved in the promotion of lateral root growth [54]. Reduced root growth angle (RGA): is one of the most important RSA traits for improving P acquisition in many soil-grown species. In maize and common bean, lower RGA (shallow roots) is associated with increased P accumulation and improved growth in P deficient soil where P is concentrated in the topsoil [55–57]. However, in aquatic systems when P is likely homogenously distributed, a lower RGA is unlikely to be advantageous. Root growth angle therefore is not considered important as a trait for enhance PUE in aquatic plants.

**Increased lateral root density:** (arising from the pericycle of mature roots) also enhances P acquisition by allowing plants to explore outside P-depleted zones. Using maize recombinant inbred lines (RILs) with contrasting lateral rooting phenotypes, significant differences in phosphorus acquisition, biomass accumulation and relative growth rate were observed under low phosphorus availability [58, 59]. Increased investment in the production of lateral roots was shown to be cost-effective under low P. For aquatic crops, enhanced lateral root density is an important trait for enhanced PUE since it increases root surface area for P uptake.

**Adventitious roots:** from above ground structures can enhance topsoil foraging by up to 10% in stratified soil [60]. These require a lower metabolic investment than basal roots, however, in uniform soil they can limit P acquisition by hindering the growth of basal roots [61]. In aquatic and flooding-tolerant plants, adventitious roots are important for P uptake within the water column [62, 63]. There is positive correlation between the size of the adventitious root system and P uptake in bittersweet (*Solana dulcamara*) plants under long-term submergence, and as mentioned, adventitious roots are responsible for a higher proportion of P acquisition in watercress than basal roots [32, 64]. Thus, in an aquatic crop such as watercress, increased adventitious root production is a key trait for enhanced PUE.

**Root cortical aerenchyma:** (RCA; enlarged spaces in the root cortex) are an adaptation to waterlogged soils and reduce the risk of asphyxiation and their formation has been suggested to increase P uptake in deficient soils [65]. Several studies have reported increase in RCA formation in maize under these conditions [66–68]. Modelling of root architecture in maize and bean has shown RCA may increase the growth of plants up to 70% in maize and 14% in bean under low P availability largely due to remobilisation of P from dying cells [67]. However, formation of aerenchyma is also associated with reduced root hydraulic conductivity in maize which may impede transport of water, but is unlikely to be relevant to hydroponically grown crops [69]. Most aquatic plants, including watercress, form aerenchyma constitutively in roots and stems to aid internal gas exchange and maintain strength under water pressure [70]. For aquatic crops such as watercress, formation of root aerenchyma is an important trait for selection for enhanced PUE.

**Root hair density and root hair length:** root hair density increases up to 5 times in low P conditions [71–73]. Using *Arabidopsis* mutants, Bates & Lynch (1996) found hairless plants had lower biomass and produced less seed than wild-type plants at low phosphorus availability [71]. Root hairs increased root surface area by 2.5 to 3.5-fold in barley and wheat (*Triticum aestivum*), respectively, and there was an almost perfect correlation between P uptake and root hair surface area [74]. Root hair traits vary substantially between genotypes and the genetic control underlying their formation is well understood, thus making them an excellent target for plant breeding programs [75–78]. Root hair length and density are likely to be important for PUE in aquatic crops as they significantly increase root surface area for P uptake.

Synergism of root phenotypes should also be considered. A modelling approach in *Arabidopsis* showed that the combined effects of root hair length, root hair density, tip to first root hair distance and number of trichoblast files (responsible for forming root hairs) on P acquisition was 3.7-fold greater than their additive effects [72]. For aquatic species such as watercress, the root ideotype for determining optimal P acquisition remains unknown. Although, absorption of P through the shoots is still debated, root uptake is generally regarded as the mode of P uptake in aquatic plants [79–81]. Watercress beds have a fine gravel substrate which contains negligible amounts of P [82]. The substrate within watercress beds is likely too shallow to allow for significant stratification of P, thus a shallower basal root angle would be unlikely to provide much adaptive benefit. In groundwater sources, P may be distributed more homogeneously due to turbulent flowing water, so RGA will not assist within the water column. Nevertheless, even with homogenous P distribution, plants with shallower root systems have been shown to encounter less inter-root competition with roots on the same plant so RGA could provide an adaptive value in this sense [83]. Cumbus & Robinson (1977) studied P absorption by the adventitious and basal roots of watercress and found that the adventitious roots absorbed a higher proportion of P at low P concentrations, despite having
a lower biomass compared to the basal root tissue [32]. Thus, adventitious roots are also a key trait for analysing watercress PUE. Increased production of lateral roots, adventitious roots and root hairs all increase root surface area, and thus will increase P acquisition from the water and sediment and are important traits for PUE in watercress.

In addition, the root cap can account for 20% of the phosphate absorbed by the roots of Arabidopsis [84]. Therefore, increasing the number of roots increases the number of root tips and the number of these "hot spots" for phosphate acquisition.

**Improving P acquisition at the molecular level: P transporters, organic acid exudation, phosphatase secretion**

Plants are reliant on phosphate transporters to acquire P from the environment and transport P between tissues, and this includes for aquatic plants. The PHT1 (PHOSPHATE TRANSPORTER 1) family is the most widely studied group of P transporters and is primarily responsible for P uptake but also has a role for P transport between tissues [85–87]. A broad range of expression patterns are associated with different PHT1 genes but generally, higher expression of PHT1 genes is associated with improved shoot biomass accumulation and P tolerance [88–90]. Watercress with higher PHT1 expression may result in improved biomass accumulation in P deficient water, but this has yet to be tested.

Additional traits that are important in other crops are organic acid (OA) exudation and phosphatase activity. Since these control release of P from organic forms in the soil, they are less relevant to watercress cultivation where P released from bound sources would be rapidly lost to the watercourse. However, phosphatases that remobilise P from intracellular sources have been identified in Arabidopsis so similar phosphatases could enhance internal P utilisation in watercress [91, 92].

**Traits for improving P utilisation**

**Improving utilisation at the molecular level**

Alongside phosphate acquisition, PUE also refers to more efficient P utilisation associated with re-translocation and recycling of stored P, that relies on effective P transport within the plant, P scavenging, and use of alternate biochemical pathways that bypass P use [93–95]. Re-translocation between plant tissues is governed by transporters such as PHT transporters and PHO transporters. Unlike, PHT transporters which regulate P acquisition too, PHO transporters are solely responsible for P transport into vascular tissues and cells [96]. The genetic control underlying these PUE mechanisms is covered in the subsequent section.

Alternative P use strategies includes substituting phospholipids in cell walls with sulfolipids and galactolipids. Several enzymes in the glycolytic pathway depend on P so bypass enzymes such as pyrophosphate-dependent phosphofructokinase (PPI-PFK), phosphofructo-

**Understanding the genetic control of PUE in watercress**

Many agricultural traits, such as PUE, are under the control of multiple rather than single genes, defined as quantitative traits and can be mapped on the genome as quantitative trait loci (QTL) and used to identify co-located candidate genes [104]. QTL mapping and candidate gene mining are well-established techniques and linking phenotype to genotype in this way provides understanding of the genetic control of PUE traits of interest. These genomic loci can then be used to develop individual molecular markers for breeding or in genomic selection strategies that utilise multiple marker data.

**QTL for PUE**

QTL for overall PUE metrics as well as QTL for more specific architectural root traits associated with low P tolerance have been identified in several economically important crops including soybean, soybean (Glycine max), rice (Oryza sativa), maize and common bean [105–109]. RSA is extremely plastic, subject to effects of hormone signalling, environmental stimuli and under the control of several genes so elucidating these QTL is challenging [110, 111].

Studies on other Brassicaceae species are likely of most genetic relevance for QTL mapping in watercress, however QTL associated with other species such as soybean, rice, sorghum and wheat are summarised in Table 1. P-starved Arabidopsis exhibit longer root hairs and higher root hair density, decreased primary root length and increased lateral root density [112, 113]. Three QTL, (including one QTL later identified as the gene LPR1) were identified which explained 52% of the variance in primary root length [114]. In rapeseed (Brassica napus) primary root length decreases, lateral root length and density increases with declining P concentration [115]. Several QTL are associated with these changes and many co-locate with QTL for root traits in Arabidopsis. A more recent study used over 13 000 SNP markers to construct a genetic linkage map in rapeseed, where 131 QTL were identified in total across different growth
systems and P availabilities \[116\]. However, only four QTL were common to all conditions, demonstrating strong environmental effects determining these QTL. To date, there is no published literature on QTL associated with aerenchyma formation under low P in any plant species and no studies exist on QTL mapping for root traits in watercress. Identification of QTL and markers associated with PUE could accelerate breeding for nutrient use and reduce the environmental impact associated with watercress cultivation.

**Candidate genes for PUE**

Genes involved in phosphate acquisition and utilisation identified in other species for PUE are likely to be candidate genes in aquatic crops like watercress. A set of 95 core phosphate starvation-inducible genes, whose expression changes $>2$ fold under P deficiency have been identified in Arabidopsis, resulting in the identification of candidate genes for PUE in several other crops including wheat, maize, oat (Avena sativa) rice and white lupin (Lupinus albus) under P deficiency \[131–136\]. The Arabidopsis “phosphatome” contains a large portion of genes whose function have not been characterised under P deprivation, and thus many of these are not discussed here. A selection of the most important PUE genes are summarised in Table 2.

**Genes for P acquisition: Morphological adaptations, PUE transcription factors and P transporters**

Specific genes involved in root architecture are targets for enhanced PUE. Although RSA traits are highly quantitative, a BLAST to the rapeseed reference genome revealed 19 candidate genes related to root growth and genetic responses to low P in Arabidopsis \[116\]. These genes included AUXIN-INDUCED IN ROOT CULTURES 12 (AIR12) involved in auxin-induced production of lateral roots and PHOSPHATE DEFICIENCY RESPONSE 2 (PDR2) which is part of growth changes in the plant apical meristem under P deficiency \[137, 138\]. PDR2 is a major component of the P starvation response and functions together

---

**Table 1. A selection of studies identifying QTL associated with various PUE traits in terrestrial species.** No studies exist for QTL mapping of PUE in aquatic crops but where studies include hydroponic growth systems this is noted.

| Species                  | QTL identified                                                                 | Details                                                                 | Reference |
|--------------------------|-------------------------------------------------------------------------------|------------------------------------------------------------------------|-----------|
| Arabidopsis thaliana     | Three QTL for primary root length                                             | LPR1: explained 52% of variance in primary root length (PRL).          | \[114\]   |
|                         |                                                                                | 2\(^{nd}\) QTL (LPR2) for primary root cell elongation under low P.      |           |
|                         |                                                                                | 3\(^{rd}\) QTL (LPR3) associated with PRL regardless of P status.       |           |
|                         |                                                                                | Identified 352 genes associated with leaf anion concentration under different P conditions. |           |
| Barley                  | 17 QTL for PUE, yield and phosphate acquisition efficiency                     | QTL explained 11–24.7% of variation. 14 candidate genes for P efficiency identified from these QTL. |           |
| Barley                  | 131 QTL across different environments and P conditions – 4 common to all     | Associated with numerous root growth and yield traits. 1 major QTL explaining 18% of variation in lateral root density. Hydroponic growth systems were included. |           |
| Barley                  | 38 QTL for RSA and biomass traits                                             | 3 loci accounted for 27.9% of variation in primary root length at low P. Many QTL co-locate with biomass QTL in other studies. |           |
| Barley                  | 71 QTL total for traits including biomass and measures of PUE                | 28 QTL were specific to low P conditions. Hydroponically grown so of interest for aquatic crops. |           |
| Bean                    | 16 QTL for RGA                                                                | Associated with gravitropic root traits but together only explain 15.9% of total variability in RGA |           |
| Bean                    | 19 QTL for adventitious root formation                                         | Two of the 19 QTL accounted for 61% of variation in adventitious root formation under low P. Includes hydroponic environment. |           |
| Rice                    | 16 QTL for several P tolerance traits                                         | Associated with numerous traits including biomass, root:shoot ratio, root volume, P content in seed. Including a QTL hotspot of 10 QTL, 5 of which were major QTL. |           |
| Rice                    | Pup1 – root growth and P tolerance                                            | Identification of PSTOL1 gene associated with this QTL. Overexpressor PSTOL1 plants had enhanced root growth, P uptake and up to 60% higher grain yield in P-deficient conditions. Included study in hydroponic systems. |           |
| Rice                    | 18 QTL for PUE and root:shoot ratio                                           | One common QTL for 3 traits (qRPUUE9.16): phosphorus use, relative physiological phosphate use efficiency, and relative phosphate uptake efficiency 4 candidate genes located on this QTL. |           |
| Rice                    | 21 QTL associated with plant growth inhibition under P deprivation           | 158 genes co-located with QTL e.g. GLYCEROPHOSPHODIESTER PHOSPHODIESTERASE 13, involved in Pi transport. |           |
| Sorghum                 | 14 QTL for grain yield/root morphology                                        | Grain yield QTL linked to three root morphology QTL. These QTL are tightly linked and near homologs of rice PSTOL1 gene. Hydroponics used for root phenotyping. |           |
| Soybean                 | 172 QTL (3 major) for P use and photosynthetically related traits under P stress | 3 major QTL (q14–2, q15–2, and q19–2), q14–2 for root dry weight and P uptake is underlined by the gene GmETO1. Included experiments using hydroponics. |           |

---
Table 2. Candidate genes for PUE breeding in watercress categorised based on function under P deprivation

| Candidate gene                                      | Function for PUE                                                                 | Mutant phenotypes                                                                 | References |
|-----------------------------------------------------|----------------------------------------------------------------------------------|-----------------------------------------------------------------------------------|------------|
| **P-responsive root development:**                   |                                                                                  |                                                                                  |            |
| PHOSPHATE DEFICIENCY RESPONSE 2 (PDR2)              | ● Involved in gene expression changes in the plant apical meristem under P-      | Pdr2 in P-:  
  ● Shorter primary root  
  ● Reduced P acquisition  | At: [139, 174] |
| LOW PHOSPHATE ROOT 1 (LPR1)                        | ● Functions together with LPR1.  
  ● Implicated in auxin responses to P starvation-mediated RSA changes.        | Lpr1 in P-:  
  ● Decreased root hair density  
  ● Increased primary root length  
  ● Higher P uptake.  | At: [140, 174, 175] |
| **PHOSPHATE DEFICIENCY ROOT HAIR DEFECTIVE1 (PE1)   | ● Role in signalling during root development.                                   | Per1 in P-:  
  ● 60% reduction root hair length  
  ● No significant differences in P content  | At: [176] |
| TRANSPORT INHIBITOR RESPONSE1 (TIR1)               | ● Involved in the lateral root response by increasing degradation of transcriptional repressors of auxin genes (AUX/IAA proteins) in P-  
  ● Functions together with LPR2 and PDR2 to alter root meristem activity.  | TIR1 in P-:  
  ● Impaired in lateral root formation = reduced root surface area and P uptake.  | At: [149] |
| AUXIN RESPONSE FACTOR 19 (ARF19)                   | ● Regulates genes involved in root hair elongation.  
  ● Involved in root hair formation in P-. | Arf19 in P-:  
  ● Reduced root hair elongation  
  ● Rsl2/6 in P-:  
  ● Reduced root hair elongation  | At: [148] |
| **PHOSPHORUS-STARVATION TOLERANCE 1 (OsPSTOL1)**    | ● Enhances early root growth in rice.                                           | Rice OE in P-:  
  ● Increased root and shoot biomass and P content by ~30%  
  ● Enhances tolerance to P-deficiency by improving P uptake.  | Rice: [179] |
| **P transport (uptake and translocation):**         |                                                                                  |                                                                                  |            |
| PHT (PHOSPHATE TRANSPORTER) genes                   | ● Responsible for uptake of P via the roots and transport between tissues.      | OE in P-:  
  ● Increases P uptake, shoot P content and biomass in rice and Arabidopsis. Effects dependent on individual PHT genes.  | At: [153, 181-183]  
  Soybean: [151, 184]  
  Maize: [86, 185]  
  Rice: [186-190]  
  Rapeseed: [154, 191]  
  At: [85, 96, 142, 150, 156, 192-194]  
  Soybean: [195]  
  Maize: [196]  
  Rice: [197] |
| PHO genes (PHO1, PHO2, PHO3)                        | PHO: controls P efflux out of cells into xylem and acquisition of P into cells  
  PHO2: regulates translocation of P from shoots to roots.  | Pho2 in P-:  
  ● Severe shoot P deficiency with stunted growth due to deficient P loading into the xylem.  |            |
| **PHOSPHATE TRANSPORTER TRAFFIC FACILITATOR (PHF1)  | Codes for an accessory protein for P uptake.  
  ● Involved in trafficking of PHT1 transporter to the plasma membrane.      | Phf1:  
  ● Impaired in P uptake = constitutively display PSR.  | At: [198] |

(Continued)
| Candidate gene | Function for PUE Mutants phenotypes | References |
|----------------|-------------------------------------|------------|
| **Key multi-functional transcriptional regulators:** | | | |
| ZINC FINGER OF ARABIDOPSIS 6 (ZAT6) | Regulates root development and PSR (e.g. anthocyanin accumulation) OE in P+: | At: [199, 200] |
| | • Increased root shoot biomass and lateral root length | | |
| | • Decrease in primary root length and number of lateral roots. | | |
| | • Overall significant increase in P uptake attributed to increase root surface area. Wrky75: | At: [199] |
| | • Reduced P uptake (due to reduced expression of PHT transporter genes) | | |
| | • More susceptible to P stress. | | |
| WRKY75 | Regulates several P starvation-induced genes (e.g. phosphatases and P transporters). | | |
| | Regulates root architecture independent of P supply. | | |
| | May have mutually synergistic effects with ZAT6: | | |
| WRKY6 and WRKY42 | Negative regulators of PSR via repression of PHO1. | At: [201] |
| HPS1 (HYPERSENSITIVE TO PHOSPHATE STARVATION 1) | Involved in sugar-mediated responses to P- by interaction with SUC2 sucrose transporter gene (in vascular tissues). | At: [146] |
| | Hps1 in P+: enhances PSR: increased expression of P transporters, overproduction of APases (ACP5) and anthocyanins, shift in allocation of P from shoots to roots, shorter lateral roots and root hairs. | | |
| PHR1 (PHOSPHATE STARVATION RESPONSE 1) and PHL1 (PHR-LIKE 1) | Global regulators of several Pi-deficiency-responsive genes. | At: [141, 202] |
| | • Involved in transducing low P signals. | | |
| | • Bind to the P1BS element in several P starvation genes. | | |
| SIZ1 | Encodes a SUMO E3 ligase that is a negative regulator of P starvation-dependent signalling. | At: [203, 204] |
| | Alters RSA through control of auxin patterning | | |
| | Spx1 and spx2 in P-: no significant differences in P content | | |
| | Spx3: lower P tolerance, reduced root systems and impaired shoot growth | | |
| | Siz1 in P-: | | |
| | • Enhanced PSR: reduction in primary root elongation, increased root:shoot biomass and root hair number and length. | | |
| | Arp6 in P+: | At: [208] |
| | • Enhanced PSR | | |
| | • Decreased P content | | |
| | Ehlh32in P+: | At: [209] |
| | • Enhanced PSR: increased root hair length, P content and anthocyanin accumulation | | |
| ETHYLENE RESPONSE FACTOR070 (ERF070) | Negative regulator of P homeostasis and root growth traits. | At: [210] |
| | Eryf070 in P-: | | |
| | • Increased number and length of lateral roots, root hair number and P content | | |
| | Overexpression of MYB62: | At: [211] |
| | • Decrease in lateral root length in P+/P-, larger root:shoot ratio under P+ | | |
| | • Increased P uptake in P+ | | |
| | • Increased anthocyanin accumulation | | |
| | • Suppression of several PSR genes | | |
| | Overexpression of AT4/IPS: reduced P content due to increased PHO2 expression | | |
| | AT4: fails to redistribute P so higher shoot P accumulation | | |
| MYB62 | Roles in RSA development, P uptake and acid phosphatase activity via gibberellic acid metabolism and signalling. | | |
| ACTIN-RELATED PROTEIN6 (ARP6) | Role for modulating responses to P- via chromatin remodelling. | | |
| BHLH32 | Negative regulator of PPCK expression (involved in P scavenging), root hair formation and anthocyanin production. | | |
| | Arp6 in P+: | At: [143, 205] |
| | • Enhanced PSR | | |
| | • Decreased P content | | |
| | Ehlh32in P+: | At: [144, 206, 207] |
| | • Enhanced PSR: increased root hair length, P content and anthocyanin accumulation | | |
| | | Rice: | | |
| | | Overexpression of MYB62: | | |
| | | • Decrease in lateral root length in P+/P-, larger root:shoot ratio under P+ | | |
| | | • Increased P uptake in P+ | | |
| | | • Increased anthocyanin accumulation | | |
| | | • Suppression of several PSR genes | | |
| | | Overexpression of AT4/IPS: reduced P content due to increased PHO2 expression | | |
| | | AT4: fails to redistribute P so higher shoot P accumulation | | |

(Continued)
Table 2. Continued

| Candidate gene | Function for PUE | Mutants phenotypes | References |
|----------------|------------------|--------------------|------------|
| OsARF16        | • Role in PSR and auxin responses to root development. | Osarf16 in P-:  
• Insensitive to P deficiency  
• Lower root:shoot ratio, lower root hair length and number of lateral roots  
• Decreased expression of several PSR genes | Rice: [213] |
| OsARF12        | • Functions in P homeostasis | Osarf mutants in P+/-:  
• Increased P concentration, upregulation of P transporter genes and APase activity, decreased number of lateral roots  
• Many other P responsive genes upregulated: e.g. OsSPX1, OsSQD2, OsTIR1. | Rice: [214] |

P utilisation (P scavenging and P-bypass enzymes):

PAP26
The predominant intracellular purple acid phosphatase in Arabidopsis.
• Recycles P from intracellular P metabolites in P-
• Expression upregulated 2-fold in P-
• Assists with P remobilisation during leaf senescence.

Pap26 in P-:
• Reduced intracellular APase activity  
• 30% decrease in shoot fresh weight  
• 50% reduction in total P content in leaves

At: [157, 158, 215]

PPC1, PPC2, PPC3, PPCK1, PPCK2

 Encode Arabidopsis PEPCs.
• Provide a metabolic bypass to ensure continued pyruvate supply to tricarboxylic acid cycle in P-

Ppc1/2/3 in P+:
• Reduced shoot biomass

At: [99, 160]

Brassica spp.: [97, 100]

MGD2, MGD3

 Encode major enzymes for galactolipid biosynthesis in P-
Phospholipids in cell membranes are substituted for non-phosphorus lipids (e.g. DGDG).

Mgd2/3 in P-:
• Reduced DGDG accumulation, fresh weight, root growth and photosynthetic activity

At: [163]

PHOSPHOETHANOLAMINE/PHOSPHOCHOLINEPHOSPHATASE 1 (PECP1)

 Encodes a phosphatase that assists in the liberation of P from phospholipids.

Pecp1 in P-:
• Shorter primary root  
• No significant differences in free P content.

At: [165, 166, 169]

PHOSPHATE STARVATION-INDUCED GENE 2 (PSR2)

 Involved in the galactolipid biosynthetic process with PECPI.  
• Assists in the liberation of P under limiting conditions.

Double mutant with pecp1 in P-:
• No altered growth phenotype  
• Reduction in choline (involved in galactolipid biosynthesis) content.

At: [168, 169]

PLDZETA1 and PLDZETA2

 Hydrolyse major phospholipids which releases DAG for galactolipid synthesis and P.
• Maintains P supply in P-
• Also involved in root elongation in P-

Pldζ1p l dζ2 in P-: decrease in primary root elongation  
Pldζ2 in P-: decreased PA accumulation

At: [170–172]

SULFOQUINOVOSYLDIACYLGlycerol2 (SQD2)

 Involved in the replacement of phospholipids with sulfolipids.

Sqd2 in P-:
• Altered lipid remodelling and impaired growth

Gdpd1 in P-:
• Decreased G3P content, P content and seedling growth.

At: [98, 216]

GDPD1

 Involved in the formation of glycerol-3-phosphate (G3P) from phospholipid products, that can be dephosphorylated to release P.

Sulfoquinovosyldiacylglycerol2 (SQD2) with LPR1 and its close paralog LPR2 as a P-sensitive checkpoint in root development by monitoring environmental P concentration, altering meristematic activity and adjusting RSA [139, 140].

Genes involved in transcriptional control are multifunctional under P deprivation; some have overlapping roles in RSA development, P signalling and P utilisation. They are discussed together here despite partial involvement in P utilisation. PHR1 (PHOSPHATE STARVATION RESPONSE 1) and PHL1 (PHR-LIKE 1) code for transcription factors that play critical roles in the control of P starvation responses [141]. PHR1 mediates expression of the microRNA miR399 which modulates the PHO2 gene, responsible for P allocation between roots and shoots and affects expression of other PSR genes such as PHT transporters [142]. SPX transcription factors (SPX1, SPX2,
SPX3, SPX4) are important negative regulators of PSR via repression of PHR [143–145]. The roles of several other transcription factor genes on RSA (such as ZAT6, WRKY75, BHLH32 and OsPTF1) and other regulatory elements (such as SIZ1 and ARF6) are summarised in Table 2 and Figure 3.

Auxin, sugars and other hormones such as cytokinins, ethylene, abscisic acid (ABA), gibberellins and strigolactones are implicated in phosphate-induced determination of RSA so genes involved in these pathways may be significant candidates [103, 146, 147]. Under low P, auxin levels increase in root hair zones and root tips. Auxin mutants such as tau1 (involved in auxin synthesis) and aux1 (involved in auxin transport) have impaired root hair growth in low P [148]. Expression of the Arabidopsis auxin receptor gene TIR1 increases under low P availability which results in increased sensitivity to auxin and production of lateral roots [149]. Mutants in auxin-inducible transcription factors (e.g. ARF19, OsARF16, OsARF12, RSL2, RSL4) also have disrupted root hair responses under low P. ROOT HAIR DEFECTIVE 6-LIKE-2 (RSL2) and ROOT HAIR DEFECTIVE 6-LIKE-4 (RSL4) are responsive to P deficiency and promote root hair initiation and elongation. ARF19 is a key transcription factor promoting auxin-dependent root hair elongation in response to low P [148]. HPS1 (HYPERSENSITIVE TO PHOSPHATE STARVATION 1) is involved in regulating the sucrose transporter SUC2 and hps1 mutants exhibit significant P-starvation responses under P-sufficient conditions [146]. Plants with impaired cytokinin receptors CRE1 and AHK3 show increased sugar sensitivity and increased expression of P-starvation genes [150]. ETHYLENE RESPONSE FACTOR070 (ERF070) is a transcription factor critical for root development under P starvation. Though no studies exist for P-associated gene expression changes in watercress, Müller et al. (2021) used RNA sequencing (RNA-seq) approaches to identify responses to submergence in watercress and found several ABA biosynthesis and catabolism genes associated with stem elongation [47]. This study provides a model for using transcriptomic approaches to explore hormone-induced morphological changes in watercress.

For P acquisition, the PHT gene family controlling P transport provides several candidate genes. In Arabidopsis, the PHT (PHOSPHATE TRANSPORTER) genes that encode phosphate transporters responsible for transport of P anions are well characterised and are grouped into four families (PHT1, PHT2, PHT3 and PHT4). PHT proteins other than PHT1 are involved in the uptake, distribution and remobilisation of P within the plant, however, PHT1 in the plasma membrane is the most important [151–153]. Phosphate stress induces expression of these
genes. However, the use of PHT transporters in plant breeding has been limited by P toxicity and other side effects of unbalanced P regulation associated with the overexpression of some transporter genes [154]. For example, OsPHT1;9 and OsPHT1;10 overexpressing rice plants have reduced biomass under high phosphate compared to wild-type plants [87]. Accessory proteins, encoded for by genes like PHOSPHATE TRANSPORTER TRAFFIC FACILITATOR (PHF1), are also important for proper functioning of P transporter genes. Homologs of PHT1 transporter genes, transcriptional factors including the SPX gene family, and genes involved in RSA determination such as PDR2 and LPR1/2 could be candidate genes for improving phosphate acquisition in watercress, but these genes have yet to be identified in aquatic crops.

**Genes for P utilisation: P re-translocation, signalling and scavenging**

The transcriptional regulation of PUE is complex: there is some overlap with genes involved in both phosphate acquisition and phosphate utilisation, such as the global transcriptional regulation by PHR1 and PHL1. Here we target genes primarily involved in utilisation, including those responsible for P transport within the plant, alternate metabolic pathways, and internal P-scavenging. Using an Arabidopsis Affymetrix gene chip, changes in global gene expression have been analysed in response to P deprivation [153]. The expression of 612 genes was induced and 254 genes suppressed including upregulation of phosphate transporters such as PHT1 genes and PHO1;H1 (involved in P loading into xylem vessels) [152]. Genes involved in protein biosynthesis were downregulated during deficiency, likely representing P recycling strategies.

PHT transporters also play a role in P utilisation through re-translocation of P within the plant. Five of the 13 maize PHT1 genes are induced in other tissues such as leaves, anthers, pollen and seeds, suggesting PHT1 involvement in diverse processes such as root-to-shoot distribution [86]. PHO1 is another central element responsible for P homeostasis and transport. Pho1 mutants exhibit P deficiency in the shoots due to lack of P loading into the xylem vessels [96, 156].

Phosphatases are important for remobilisation of fixed P. Eleven genes encoding different purple acid phosphatases were reported to be upregulated under P starvation in Arabidopsis [152]. The Arabidopsis genome encodes 29 purple acid phosphatases, some of which are excreted into the soil, such as PAP12 and PAP26 [92, 157]. Only phosphatase activity within the plant is relevant to watercress breeding: in a flowing water system, any P made available around the roots by secreted phosphatases would rapidly wash away. As well as being a major secreted phosphatase, PAP26 is regarded as the predominant intracellular acid phosphatase in Arabidopsis and is upregulated two-fold under P-deficiency [158]. PAP26 functions in PUE by scavenging P from intracellular and extracellular P-ester pools, to increase P availability in the plant. Homologs of PAP26 should be investigated in watercress.

Plants also respond to P starvation by utilising alternative metabolic pathways. Genes involved in lipid metabolism and biosynthesis represent the largest group of core PSI (phosphate starvation inducible) genes in Arabidopsis, demonstrating their importance in PUE under P starvation [133]. Three genes encode “plant-type” PEPCK enzymes in Arabidopsis (PPC1, PPC2, PPC3). PPC1 is expressed in roots and flowers, PPC2 in all organs and PPC3 in only roots [159, 160]. PEPCK activity is affected by phosphorylation by PPCK (PEPC kinase), thus PPCK1 and PPCK2 genes are additional important components for P-bypassing [99, 161].

Membrane phospholipids constitute approximately 20% of the total P in the leaves of P-sufficient plants [162]. This represents a large pool of P that can be remobilised. 7% of the P-responsive genes found by Misson et al. (2005) were involved in lipid biosynthesis pathways in Arabidopsis. This includes the genes MGD2 and MGD3 whose expression changed 11-fold and 48-fold under P deficiency, respectively. These genes encode major enzymes for galactolipid biosynthesis and are involved in replacing phospholipids in cell membranes with non-phosphorus lipids such as galactolipid digalactosyldiacylglycerol (DGDG) [163]. PECP1 is involved in the liberation of P from phospholipids and is upregulated under P deprivation, with up to 1785-fold increases in expression reported in roots [164–166]. PSR2 encodes a phosphatase involved in galactolipid biosynthesis and whose expression increases 174-fold in P deprived seedlings [167]. Both PECP1 and PSR2 have similar roles in the dephosphorylation of phosphocholine (PCho) in the galactolipid synthesis pathway. However, despite their massive upregulation, it has been observed that inactivation of PECP1 and PSR2 does not alter plant growth or plant P content under P-deprivation so PCho is not likely a major source of P under limiting conditions [168, 169]. PLDζ1 and PLDζ2 encode phospholipases D zeta 1 and 2 that hydrolyse major phospholipids such as phosphatidylcholine which yields phosphatidic acid (PA) and PA phosphatase (PAP) and releases DAG (for galactolipid synthesis) and P [170–172]. Phospholipids can also be replaced by sulfolipids. SQD2 is the primary gene in this pathway and encodes an enzyme that catalyses the final step in the sulfolipid biosynthesis [98]. GDPD1 is involved in the formation of glycerol-3-phosphate (G3P) from phospholipid products (such as glycerolphosphoglycerol), that can be dephosphorylated to release P [173].

Homologs of PHT1 genes responsible for P redistribution, within the plant, genes involved in P scavenging (i.e. PAP26), genes implemented in metabolic pathways that bypass P use including galactolipid biosynthetic pathways (e.g. MGD2/3, PLDζ1/2, PECP1 and PSR2) and those involved in sulfolipid biosynthesis (e.g. SQD2) could be candidate genes for improving phosphate utilisation in watercress.
Status of watercress breeding

Watercress root research is virtually completely absent in the literature, with no studies on root responses to phosphate availability. Nevertheless, the finite nature of rock phosphate and the fact that watercress cultivation methods have the potential to result in environmental damage (associated with fertiliser input to waterways), are clear drivers, as with soil-grown crops, to breed for watercress with improved PUE. Uncovering phenotypic traits and the molecular basis for PUE are important early steps in breeding for phosphate use efficiency.

No commercial breeding programs exist for watercress worldwide, but germplasm collections are emerging [217–219]. Development of new watercress varieties is also no doubt limited by the lack of genomic information for this crop. Payne (2011) screened a germplasm collection under indoor and outdoor conditions and identified significant variation in several traits including stem length, stem diameter and antioxidant concentrations [218]. Voutsina (2017) used RNA-seq to analyse the first watercress transcriptome and identified differences in antioxidant capacity and glucosinolate biosynthesis across a germplasm collection of watercress, with 71% of the watercress transcripts annotated based on orthology to Arabidopsis [220]. Jeon et al. (2017) independently used RNA-seq approaches to assemble the watercress transcriptome de novo. They identified 33 candidate genes related to glucosinolate biosynthetic pathways using Arabidopsis glucosinolate genes to search for homologous sequences in the watercress transcriptome [221].

Additionally, a watercress mapping population comprising 259 F2 individuals was established by Voutsina (2017) using parents with contrasting nutrient and growth phenotypes. Genotype-by-sequencing of this mapping population enabled the construction of first genetic linkage map for watercress and identified 17 candidate genes related to glucosinolate biosynthesis. Together these findings will assist in developing commercial cultivars with a reduced need for phosphate, and a reduced negative environmental impact.

To assess whether homologs of the candidate PUE genes identified in this study exist in watercress, available watercress transcriptome data was mined for 13 key PUE genes selected from Table 2. This included genes whose expression was induced more than tenfold in at least 2 independent studies (as found by (Lan et al., 2015), plus PHR1, the global regulator of P starvation responses. Annotated transcripts were obtained from transcriptomic studies of watercress by Voutsina et al., 2016; Jeon et al., 2017; Müller et al., 2021 and matches for these candidate genes were assessed by searching for genes using AGI (Arabidopsis Gene Identifiers) [47, 133, 221, 222]. Across all studies, strong matches were found for PHT1;4, SPX1, PHR1, MDG3, PEP1, PLD1/2, PSR2 and SQD2, with all corresponding e-values ranging from 0 to 3.00E-32. Additionally, Müller et al. identified homologous transcripts for MDG2. Voutsina et al. had transcripts corresponding to PHT1;3, and MDG2, and Jeon et al. had hits for PHT1;2 and PHT1;3. No matching transcripts were found for PHT1;1 in any of the three studies. Where FDR values (p-values) were < 0.05, changes to expression patterns were noted. Interestingly, Müller et al. also observed varying levels of upregulation of PHT1;4 and PHR1 following submergence, which may suggest a link between phosphate starvation and submergence responses.

Conclusions

Watercress is a non-model leafy green crop, grown using traditional aquatic systems across the world, but also increasingly seen as a suitable crop for vertical hydroponic indoor systems. It is ranked as the top “powerhouse” food, with the highest nutrient density, including high concentrations of essential vitamins, minerals and phytonutrients [223]. However, regulation from environmental agencies on nutrient inputs to water systems is increasing the pressure to reduce fertiliser use in traditional aquatic growing systems [38, 224].

Breeding new varieties with improved phosphate use efficiency (PUE) is thus a priority for both traditional outdoor and indoor systems. Prebreeding for PUE in aquatic crops such as watercress should target a suite of traits (Fig. 3), including (i) increased total root length, (ii) number of root tips, (iii) lateral and adventitious root number and length as important root architectural traits. Microscopic traits of interest include (iv) increased aerenchyma formation, (v) increased root hair density and length. At the genomic level, (vi) enhanced activity of P transporter genes (e.g. PHT homologs) and (vii) genes involved in P utilisation, particularly those with roles in P remobilisation (e.g. PAP26, MGD3, PEPC1) and P-bypass enzymes (e.g. PPC1), may prove good candidate genes for PUE.

Barriers for root trait breeding include the difficulties in evaluating roots in the substrate, their phenotypic plasticity in response to numerous environmental factors and limited genomic knowledge for watercress. Recent developments in high-throughput phenotyping and whole-genome sequencing-based genotyping will accelerate QTL identification, the discovery of individual genes involved in PUE and lay the foundations for development of new watercress varieties with improved low P tolerance.
Acknowledgements
This work was supported by a PhD studentship funded by Vitacress Salads Ltd to LH and by the John B Orr endowed chair in Environmental Plant Sciences held by GT at the University of California, Davis.

Author contributions
LH conducted the literature review, made figures, and drafted the manuscript. GT conceived the study and led on the project, provided comments and modified the manuscript. All authors have reviewed and approved final submission.

Conflict of interests
The authors declare no competing interests.

References
1. Zeven AC, Zhukovsky PM. Dictionary of Cultivated Plants and their Centres of Diversity. Pudoc; 1975.
2. Casey H, Smith SM. The effects of watercress growing on chalk headwater streams in Dorset and Hampshire. Environ Pollut. 1994;85:217–28.
3. Berrie AD. The chalk-stream environment. Hydrobiologia. 1992;248:3–9.
4. Schindler DW, Hecky RE, Findlay DL et al. Eutrophication of lakes cannot be controlled by reducing nitrogen input: results of a 37-year whole-ecosystem experiment. Proc Natl Acad Sci. 2008;105:11254–8.
5. Cox J. Watercress Growing and its Environmental Impacts on Chalk Rivers in England. https://publications.naturalengland.org.uk/publication/40010 (2009).
6. Schachtman DP, Reid RJ, Ayling SM. Phosphorus uptake by plants: from soil to cell. Plant Physiol. 2008;146:265–79.
7. Oerke EC. Crop yields: trends and future potential. Annu Rev Energy Environ. 2003;28:1–46.
8. Chase T, North P, suitcase L et al. A review of the watercress growing environment. J Environ Qual. 2011;40:1993–2000.
9. Scholes RJ, Archer S. Soil carbon cycle responses to environmental change. Annu Rev Ecol Evol Syst. 2001;32:485–512.
10. Hoppe SD, Elliott DE, Reuter DJ. Plant tests for diagnosing phosphorus deficiency in barley (Hordeum vulgare L.). Aust J Exp Agric. 1999;39:855–68.
11. Lynch J, Läuchli A, Epstein E. Vegetative growth of the common bean in response to phosphorus nutrition. Crop Sci. 1991;31:380–7.
12. Steen I. Phosphorus availability in the 21st century: management of a non-renewable resource. Phosphorus Potassium. 1998;217:1–13.
13. Smil V. Phosphorus in the environment: natural flows and human interferences. Annu Rev Energy Environ. 2000;25:53–88.
14. Fao F. Use of phosphate rocks for sustainable agriculture. http://www.fao.org/3/y5053en/y5053e00.htm#Contents (2004).
15. Rosemarin A, Ekane N. The governance gap surrounding phosphorus. Fertilizer research. 2016;104:265–79.
16. USGS. Phosphate Rock Statistics and Information. https://www.usgs.gov/centers/nmic/phosphate-rock-statistics-and-information (2019).
17. Cordell D, Drangert J-O, White S. The story of phosphorus: global food security and food for thought. Glob Environ Change. 2009;19:292–305.
18. Schaum C. Phosphorus: polluter and resource of the future. IWA Publishing. 2018.
19. Oberle B, Stefan B, Steve H-D et al. Global Resources Outlook 2019. https://www.resourcepanel.org/reports/global-resources-outlook (2019).
20. MacDonald GK, Bennett EM, Potter PA et al. Agronomic phosphorus imbalances across the world’s croplands. Proc Natl Acad Sci. 2011;108:3086–91.
21. Hedley MJ, Stewart JW, Chauhan BS. Changes in inorganic and organic soil phosphorus fractions induced by cultivation practices and by laboratory incubations. Soil Sci Soc Am J. 1982;46:970–6.
22. Hou E, Tan X, Heenan M et al. A global dataset of plant available and unavailable phosphorus in natural soils derived by Hedley method. Sci Data. 2018;5:1–13.
23. Harrison AF. Soil Organic Phosphorus: A Review of World Literature. CAB International; 1987.
24. Mclaughlin MJ, Baker TG, James TR et al. Distribution and forms of phosphorus and aluminium in acidic topsoils under pastures in south-eastern Australia. Soil Res. 1990;28:371–85.
25. Shand CA, Macklon AES, Edwards AC et al. Inorganic and organic P in soil solutions from three upland soils. Plant Soil. 1994;159:255–64.
26. Reddy KR, Wang Y, DeBusk WF et al. Forms of soil phosphorus in selected hydrologic units of the Florida Everglades. Soil Sci Soc Am J. 1998;62:1134–47.
27. Lindsay WL. Chemical Equilibria in Soils. Wiley; 1979.
28. Frossard E, Condron LM, Oberson A et al. Processes governing phosphorus availability in temperate soils. J Environ Qual. 2000;29:15–23.
29. Hinsinger P. Bioavailability of soil inorganic P in the rhizosphere as affected by root-induced chemical changes. Review. 2001;237:173–95.
30. Baldwin DS, Beattie JK, Coleman LM et al. Phosphate ester hydrolysis facilitated by mineral phases. Environ Sci Technol. 1995;29:1706–9.
31. Reynolds CS, Davies PS. Sources and bioavailability of phosphorus fractions in freshwater: a British perspective. Biol Rev. 2001;76:27–64.
32. Cumbus IP, Robinson LW. The function of root systems in mineral nutrition of watercress (Rorippa nasturtium-aquaticum (L) Hayek). Plant Soil. 1977;47:395–406.
33. Dawson CJ, Hilton J. Fertiliser availability in a resource-limited world: production and recycling of nitrogen and phosphorus. Food Policy. 2011;36:14–22.
34. Stutter MI, Jackson-Blake L, May L et al. Factoring Ecological Significance of Sources into Phosphorus Source Apportionment. https://www.crew.ac.uk/sites/www.crew.ac.uk/files/sites/default/files/publication/Factoring%20Ecological%20Significance%20of%20Sources%20into%20Phosphorus%20%20%20%20%20Source%20Apportionment.pdf (2014).
35. Withers PJ, Jordan P, May L et al. Do septic tank systems pose a hidden threat to water quality? Front Ecol Environ. 2014;12:123–30.
36. Meekonnen MM, Hoekstra AY. Global anthropogenic phosphorus loads to freshwater and associated grey water footprints.
and water pollution levels: a high-resolution global study. Water Resour Res. 2018;54:345–58.

37. European Environment Agency. European Waters – Assessment of Status and Pressures 2018. https://www.eea.europa.eu/publications/state-of-water (2018).

38. European Environment Agency. The European Environment — State and Outlook 2020. https://www.eea.europa.eu/publications/soer-2020 (2019).

39. Dodds WK, Bouska WW, Eitzmann JL et al. Eutrophication of U.S. freshwaters: analysis of potential economic damages. Environ Sci Technol. 2009;43:12–9.

40. Akhtar MS, Oki Y, Adachi T et al. Relative phosphorus utilization efficiency, growth response, and phosphorus uptake kinetics of brassica cultivars under a phosphorus stress environment. Commun Soil Sci Plant Anal. 2007;38:1061–85.

41. Wang X, Shen J, Liao H. Acquisition or utilization, which is more critical for enhancing phosphorus efficiency in modern crops? Plant Sci. 2010;179:302–6.

42. Parentoni SN, de Souza Junior CL. Phosphorus acquisition and internal utilization efficiency in tropical maize genotypes. Pesq agropec bras. 2008;43:893–901.

43. Shenoy VV, Kalagudi GM. Enhancing plant phosphorus use efficiency for sustainable cropping. Biotechnol Adv. 2005;23:501–13.

44. Hammond JP, Broadley MR, White PJ et al. Shoot yield drives phosphorus use efficiency in Brassica oleracea and correlates with root architecture traits. J Exp Bot. 2009;60:1953–68.

45. Neto AP, Favarin JL, Hammond JP et al. Analysis of phosphorus use efficiency traits in Coffea genotypes reveals Coffea arabica and Coffea canephora have contrasting phosphorus uptake and utilization efficiencies. Front Plant Sci. 2016;7:1–10.

46. Cesaroni I, Ioio RD, Kirschner GK et al. Plant science’s next top models. Ann Bot. 2020;126:1–23.

47. Müller JT, van Veen H, Bartylla MM et al. Relative phosphorus acquisition efficiency in maize (Zea mays) seedlings. Funct Plant Biol. 2004;31:949–58.

48. Cesarino I, Oki Y, Adachi T et al. Relative phosphorus utilization efficiency, growth response, and phosphorus uptake kinetics of brassica cultivars under a phosphorus stress environment. Commun Soil Sci Plant Anal. 2007;38:1061–85.

49. Hammond JP, Broadley MR, White PJ et al. Shoot yield drives phosphorus use efficiency in Brassica oleracea and correlates with root architecture traits. J Exp Bot. 2009;60:1953–68.

50. Grand View Research. Indoor Farming Market Size & Share Report, 2021–2028. https://www.grandviewresearch.com/industriy-analysis/indoor-farming-market (2021).

51. Lynch JP. Root architecture and plant productivity. Plant Physiol. 1995;109:7–13.

52. Gruber BD, Giehl RFH, Friedel S et al. Plasticity of the Arabidopsis root system under nutrient deficiencies. Plant Physiol. 2013;163:161–79.

53. Poorter H, Ryser P. The limits to leaf and root plasticity: what is so special about specific root length? New Phytol. 2019;206:1188–90.

54. Fromm H. Root plasticity in the pursuit of water. Plan Theory. 2019;8:1–19.

55. Zhang Z, Liao H, Lucas WJ. Molecular mechanisms underlying phosphate sensing, signaling, and adaptation in plants. J Integr Plant Biol. 2014;56:192–220.

56. Bonser AM, Lynch J, Snapp S. Effect of phosphorus deficiency on growth angle of basal roots in Phaseolus vulgaris. New Phytol. 1996;132:281–8.

57. Zhu J, Kaeppler SM, Lynch JP. Topsoil foraging and phosphorus acquisition efficiency in maize (Zea mays). Funct Plant Biol. 2005;32:749–62.

58. Malamy JE, Benfey PN. Organization and cell differentiation in lateral roots of Arabidopsis thaliana. Development. 1997;124:33–44.

59. Zhu J, Lynch JP. The contribution of lateral root to phosphorus acquisition efficiency in maize (Zea mays) seedlings. Funct Plant Biol. 2004;31:949–58.

60. Miller CR, Ochoa I, Nielsen KL et al. Genetic variation for adventitious rooting in response to low phosphorus availability: potential utility for phosphorus acquisition from stratified soils. Funct Plant Biol. 2003;30:973–85.

61. Walk TC, Jaramillo R, Lynch JP. Architectural tradeoffs between adventitious and basal roots for phosphorus acquisition. Plant Soil. 2006;279:347–66.

62. Bristow JM, Whitcombe M. The role of roots in the nutrition of aquatic vascular plants. Am J Bot. 1971;58:8–13.

63. Steffens B, Rasmussen A. The physiology of adventitious roots. Plant Physiol. 2016;170:603–17.

64. Zhang Q, Huber H, SJM B et al. Benefits of flooding-induced aquatic adventitious roots depend on the duration of submergence: linking plant performance to root functioning. Ann Bot. 2017;120:171–80.

65. Jackson MB, Armstrong W. Formation of aerenchyma and the processes of plant ventilation in relation to soil flooding and submergence. Plant Biol. 1999;1:274–87.

66. Drew MC, He CJ, Morgan PW. Decreased ethylene biosynthesis, and induction of aerenchyma, by nitrogen- or phosphate-starvation in adventitious roots of Zea mays L. Plant Physiol. 1989;91:266–71.

67. Postma JA, Lynch JP. Root cortical aerenchyma enhances the growth of maize on soils with suboptimal availability of nitrogen, phosphorus, and potassium. Plant Physiol. 2011;156:1190–201.

68. Diaz AS, Aguiaar GM, Pereira MP et al. Aerenchyma development in different root zones of maize genotypes under water limitation and different phosphorus nutrition. Biol Plant. 2018;62:561–8.

69. Fan M, Bai R, Zhao X et al. Aerenchyma formed under phosphorus deficiency contributes to the reduced root hydraulic conductivity in maize roots. J Integr Plant Biol. 2007;49:598–604.

70. Jung J, Lee SC, Choi H-K. Anatomical patterns of aerenchyma in aquatic and wetland plants. J Plant Biol. 2008;51:428–39.

71. Bates TR, Lynch JP. Stimulation of root hair elongation in Arabidopsis thaliana by low phosphorus availability. Plant Cell Environ. 1996;19:529–38.

72. Zu B, Bielenberg DG, Brown KM et al. Relative phosphorus acquisition efficiency in maize (Zea mays) seedlings. Funct Plant Biol. 2004;31:949–58.

73. Müller M, Schmidt W. Environmentally induced plasticity of root hair development in Arabidopsis. Plant Physiol. 2004;134:409–19.

74. Gahoonia TS, Care D, Nielsen NE. Root hairs and phosphorus acquisition of wheat and barley cultivars. Plant Soil. 1997;191:181–8.

75. Datta S, Kim CM, Pernas M et al. Root hairs: development, growth and evolution at the plant-soil interface. Plant Soil. 2011;346:1–14.

76. Grierson CS, Parker JS, Kemp AC. Arabidopsis genes with roles in root hair development. J Plant Nutr Soil Sci. 2001;164:131–40.
77. Lynch JP. Root phenes for enhanced soil exploration and phosphorus acquisition: tools for future crops. Plant Physiol. 2011;156:1041–9.

78. Rongsawat T, Peltier J-B, Boyer J-C et al. Looking for root hairs to overcome poor soils. Trends Plant Sci. 2021;26:83–94.

79. Barko JW, Gunnison D, Carpenter SR. Sediment interactions with submersed macrophyte growth and community dynamics. Aquat Bot. 1991;41:41–65.

80. Brix H, Lynghy JE. Uptake and translocation of phosphorus in eelgrass (Zostera marina). Mar Biol. 1985;90:111–6.

81. Christiansen NH, Andersen FØ, Jensen HS. Phosphate uptake kinetics for four species of submersed freshwater macrophytes measured by a 33P phosphate radioisotope technique. Aquat Bot. 2016;128:58–67.

82. Crisp DT. Input and output of minerals for a small watercress bed fed by chalk water. J Appl Ecol. 1970;7:117–40.

83. Lynch JP, Brown KM. Topsoil foraging – an architectural adaptation of plants to low phosphorus availability. Plant Soil. 2001;237:225–37.

84. Kanno S, Arrighi JF, Chiarenza S et al. A novel role for the root cap in phosphate uptake and homeostasis. Elife. 2016;5:1–16.

85. Hamburger D, Rezzonico E, MacDonald-Comber Petétot J et al. Identification and characterization of the Arabidopsis PHO1 gene involved in phosphate transport to the xylem. Plant Cell. 2002;14:889–902.

86. Nagy R, MJV V, Zhao S et al. Identification of quantitative trait loci for phosphorus uptake and use: critical adaptations by plants for securing a nonrenewable resource. New Phytol. 2003;157:423–47.

87. Wang X, Wang Y, Piñeros MA et al. A novel role for the root cap in phosphate uptake and homeostasis. Elife. 2016;5:1–16.

88. Wang X, Wang Y, Piñeros MA et al. Phosphate transporters from maize (Zea mays L.) under different phosphorus levels at two sites. Front Agric China. 2010;5:1786–99.

89. Vance CP, Uhde-Stone C, Allan DL. Phosphorus acquisition and use: critical adaptations by plants for securing a nonrenewable resource. New Phytol. 2003;157:423–47.

90. del Pozo JC, Allona I, Rubio V et al. A type 5 α-phosphatase gene from Arabidopsis thaliana is induced by phosphate starvation and by some other types of phosphate mobilising/oxidative stress conditions. Plant J. 1999;19:579–89.

91. Chiou T-J, Lin S-I. Signaling network in sensing phosphate availability in plants. Annu Rev Plant Biol. 2011;62:185–206.

92. Theodorou ME, Cornel FA, Duff SM et al. Phosphate starvation-inducible synthesis of the alpha-subunit of the pyrophosphate-dependent phosphofructokinase in black mustard suspension cells. J Biol Chem. 1992;267:21901–5.

93. Theodorou ME, Plaxton WC. Purification and characterization of pyrophosphate-dependent phosphofructokinase from phosphate-starved Brassica nigra suspension cells. Plant Physiol. 1996;112:343–51.

94. Plaxton WC, Tran HT. Metabolic adaptations of phosphate-starved plants. Plant Physiol. 2011;156:1006–15.

95. Wang Y, Ribot C, Rezzonico F et al. Structure and expression profile of the Arabidopsis PHO1 gene family indicates a broad role in inorganic phosphate homeostasis. Plant Physiol. 2004;135:400–11.

96. Reymond M, Svistoonoff S, Loudet O et al. Identification of QTL controlling root growth response to phosphate starvation in Arabidopsis thaliana. Plant Cell Environ. 2006;29:115–25.
115. Shi L, Shi T, Broadley MR et al. High-throughput root pheno-typing screens identify genetic loci associated with root archi-tectural traits in Brassica napus under contrasting phosphate availabilities. Ann Bot. 2013;112:381–9.

116. Zhang Y, Thomas CL, Xiang J et al. QTL meta-analysis of root traits in Brassica napus under contrasting phosphorus supply in two growth systems. Sci Rep. 2016;6:1–12.

117. El-Soda M, Moreira CN, Matongera NG et al. QTL and candidate genes associated with leaf anion concentrations in response to phosphate supply in Arabidopsis thaliana. BMC Plant Biol. 2019;19:410.

118. Gao S, Xia J, Yuan S et al. Novel QTL conferring phosphorus acquisition and utilization efficiencies in barley. Front Genet. 2020;11:1039.

119. Yang M, Ding G, Shi L et al. Detection of QTL for phosphorus effi-ciency at vegetative stage in Brassica napus. Plant Soil. 2011;339:97–111.

120. Han Y, Liao G, Shi L et al. Genetic mapping of basal root gravitropism and phosphorus acquisition efficiency in common bean. Funct Plant Biol. 2004;31:959–70.

121. Ochoa IE, Blair MW, Lynch JP. QTL analysis of adventitious root formation in common bean under contrasting phosphorus availability. Crop Sci. 2006;46:1609–21.

122. Kale RR, CHV DR, Anila M et al. Novel major QTLs associated with low soil phosphorus tolerance identified from the Indian rice landrace. PLoS I. 2021;16:e0254526.

123. Chinn JH, Gamuyao R, Dalid C et al. Developing rice with high yield under phosphorus deficiency: Pup1 sequence to applica-tion. Plant Physiol. 2011;156:1202–16.

124. Gamuyao R, Chinn JH, Paraisca-Tanaka J et al. The protein kinase Pstol1 from traditional rice confers tolerance of phosphorus deficiency. Nature. 2012;488:535–9.

125. Wissuwa M, Yano M, Ae N. Mapping of QTLs for phosphorus-deficiency tolerance in rice (Oryza sativa L.). Theor Appl Genet. 1998;97:777–83.

126. To HTM, Le KQ, Van Nguyen H et al. A genome-wide association study reveals the quantitative trait locus and candidate genes that regulate phosphorus efficiency in a Vietnamese rice collection. Physiol Mol Biol Plants. 2020;26:2267–81.

127. Mai NTP, Mai CD, Van Nguyen H et al. Discovery of new genetic determinants of morphological plasticity in rice roots and shoots under phosphate starvation using GWAS. J Plant Physiol. 2021;257:153340.

128. Bernardino KC, Pastina MM, Menezes CB et al. The genetic architecture of phosphorus efficiency in sorghum involves pleiotropic QTL for root morphology and grain yield under low phosphorus availability in the soil. BMC Plant Biol. 2019;19:87.

129. Li H, Yang Y, Zhang H et al. A genetic relationship between phosphorus efficiency and photosynthetic traits in soybean as revealed by QTL analysis using a high-density genetic map. Front Plant Sci. 2016;7:924.

130. Zhang H, Yang Y, Sun C et al. Up-regulating GmETOH1 improves phosphorus uptake and use efficiency by promoting root growth in soybean. Plant Cell Environ. 2020;43:2080–94.

131. O’Rourke JA, Yang SS, Miller SS et al. An RNA-seq transcriptome analysis of orthophosphate-deficient white lupin reveals novel insights into phosphorus acclimation in plants. Plant Physiol. 2013;161:705–24.

132. Secco D, Jaboume N, Walker H et al. Spatio-temporal transcript profiling of rice roots and shoots in response to phosphate starvation and recovery. Plant Cell. 2013;25:4285–304.

133. Lan P, Li W, Schmidt W. ‘Omic’ approaches towards understanding plant phosphorus acquisition and use. Annual Plant Reviews Volume 48: Phosphorus Metabolism in Plants. 2015;48:65–97. https://doi.org/10.1002/9781118958841.ch3.

134. Wang Y, Lyssee E, Armarego-Marriott T et al. Transcriptome and metabolome analyses provide insights into root and root-released organic anion responses to phosphorus deficiency in oat. J Exp Bot. 2018;69:3759–71.

135. Yu P, Wang C, Baldauf JA et al. Root type and soil phosphate determine the taxonomic landscape of colonizing fungi and the transcriptome of field-grown maize roots. New Phytol. 2018;217:1240–53.

136. Wang J, Qin Q, Pan J et al. Transcriptome analysis in roots and leaves of wheat seedlings in response to low-phosphorus stress. Sci Rep. 2019;9:1–12.

137. Preger V, Tango N, Marchand C et al. Auxin-responsive genes AIR12 code for a new family of plasma membrane b-type cytochromes specific to flowering plants. Plant Physiol. 2009;150:606–20.

138. Naumann C, Müller J, Sakonwasee S et al. The local phosphate deficiency response activates endoplasmic reticulum stress-dependent autophagy. Plant Physiol. 2019;179:660–76.

139. Ticconi CA, Delatorre CA, Lahner B et al. Arabidopsis pdr2 reveals a phosphate-sensitive checkpoint in root development. Plant J. 2004;37:801–14.

140. Wang X, Du G, Wang X et al. The function of LPR1 is controlled by an element in the promoter and is independent of SUMO E3 ligase SIZ1 in response to low pi stress in Arabidopsis thaliana. Plant Cell Physiol. 2010;51:880–94.

141. Bustos R, Castrillo G, Linhares F et al. A central regulatory system largely controls transcriptional activation and repression responses to phosphate starvation in Arabidopsis. PLoS Genet. 2010;6:e1001102.

142. Furi R, Pant BD, Stitt M et al. PHO2, microRNA399, and PHR1 define a phosphate-signaling pathway in plants. Plant Physiol. 2006;141:988–99.

143. Puga MI, Mateos I, Charukses R et al. SX1 is a phosphate-dependent inhibitor of PHOSPHATE STARVATION RESPONSE 1 in Arabidopsis. Proc Natl Acad Sci. 2014;111:14947–52.

144. Zhou Z, Wang Z, Ly Q et al. SX1 proteins regulate pi homeostasis and signaling in different subcellular level. Plant Signal Behav. 2015;10:1–3.

145. Qi W, Manfield IW, Muench SP et al. Developing rice with high phosphorus efficiency using rice leaf transcriptome data. Sci Rep. 2016;6:117216.

146. Lei M, Liu Y, Zhang B et al. Transcriptome analysis in roots and shoots of wheat seedlings in response to low-phosphorus solution. Plant Physiol. 2016;156:167–86.

147. Leit M, Liu Y, Zhang B et al. Genetic and genomic evidence that sucrose is a global regulator of plant responses to phosphate starvation in Arabidopsis. Plant Physiol. 2011;156:1116–30.

148. Müller R, Morant M, Jarmer H et al. Genome-wide analysis of the Arabidopsis leaf transcriptome reveals interaction of phosphate and sugar metabolism. Plant Physiol. 2007;143:156–71.

149. Bhosale R, Giri J, Pandey BK et al. A mechanistic framework for auxin dependent Arabidopsis root hair elongation to low external phosphate. Nat Commun. 2018;9:1–9.

150. Perez-Torres CA, Lopez-Bucio J, Cruz-Ramirez A et al. Phosphate availability alters lateral root development in Arabidopsis by modulating auxin sensitivity via a mechanism involving the TIR1 auxin receptor. Plant Cell. 2008;20:3258–572.

151. Fan C, Wang X, Hu R et al. The pattern of phosphate transporter 1 genes evolutionary divergence in Glycine max L. BMC Plant Biol. 2013;13:1–16.
et al. Arabidopsis thaliana PECP1: and phosphoethanolamine content. Plant Physiol. 2018; 176: 2943–62.

170. Cruz-Ramirez A, Oropeza-Aburto A, Razo-Hernandez F et al. Phospholipase D22 plays an important role in extracellular galactolipid biosynthesis and phosphate recycling in Arabidopsis roots. Proc Natl Acad Sci. 2006; 103: 6765–70.

171. Li M, Qin C, Welti R et al. Double knockouts of phospholipases D\textsubscript{1} and D\textsubscript{2} in Arabidopsis affect root elongation during phosphate-limited growth but do not affect root hair patterning. Plant Physiol. 2006; 140: 761–70.

172. Qin C, Wang X. The Arabidopsis phospholipase D family. Characterization of a calcium-independent and phosphatidylcholine-selective PLD\textsubscript{1} with distinct regulatory domains. Plant Physiol. 2002; 128: 1057–68.

173. Cheng Y, Zhou W, El Sheery NI et al. Characterization of the Arabidopsis glycerophosphodiester phosphodiesterase (GDP) family reveals a role of the plastid-localized AtGDPD1 in maintaining cellular phosphate homeostasis under phosphate starvation. Plant J. 2011; 66: 781–95.

174. Picton CA, Lucero RD, Sakhonwasee S et al. ER-resident proteins PDR2 and LPR1 mediate the developmental response of root meristems to phosphate availability. Proc Natl Acad Sci. 2009; 106: 14174–9.

175. Svistoonoff S, Creff A, Reynond M et al. Root tip contact with low-phosphate media reprograms plant root architecture. Nat Genet. 2007; 39: 792–6.

176. Li W-F, Perry PJ, Prafulla NN et al. Ubiquitin-specific protease 14 (UBP14) is involved in root responses to phosphate deficiency in Arabidopsis. Mol Plant. 2010; 3: 212–23.

177. Han Y, Xin M, Huang K et al. Altered expression of TaRSL4 gene by genome interplay shapes root hair length in allopolyploid wheat. New Phytol. 2016; 209: 721–32.

178. Yi K, Menand B, Bell E et al. A basic helix-loop-helix transcription factor controls cell growth and size in root hairs. Nat Genet. 2010; 42: 264–7.

179. Yi K, Wu Z, Zhou J et al. OsPTF1, a novel transcription factor involved in tolerance to phosphate starvation in rice. Plant Physiol. 2005; 138: 2087–96.

180. Chithrameenal K, Alagarasan G, Raveendran M et al. Genetic enhancement of phosphorus starvation tolerance through marker assisted introgression of OsPSTOL1 gene in rice genotypes harbouring bacterial blight and blast resistance. Plas One. 2018; 13: e0204144.

181. Lapis-Gaza HR, Jost R, Finnegnan PM. Arabidopsis PHOSPHATE TRANSPORTER1 genes PHT1,8 and PHT1,9 are involved in root-to-shoot translocation of orthophosphate. BMC Plant Biol. 2014; 14: 1–19.

182. Nussaume L, Kanno S, Javot H et al. Phosphate import in plants: focus on the HFT1 transporters. Front Plant Sci. 2011; 2: 1–12.

183. Remy E, Cabrito TR, Batista RA et al. The Pht1,9 and Pht1,8 transporters mediate inorganic phosphate acquisition by the Arabidopsis thaliana root during phosphorus starvation. New Phytol. 2012; 195: 356–71.

184. Qin L, Zhao J, Tian J et al. The high-affinity phosphate transporter GmPT5 regulates phosphate transport to nodules and nodulation in soybean. Plant Physiol. 2012; 159: 1634–43.

185. Liu F, Xu Y, Jiang H et al. Systematic identification, evolution and expression analysis of the Zea mays PHT1 gene family reveals several new members involved in root colonization by arbuscular mycorrhizal fungi. Int J Mol Sci. 2016; 17: 1–18.

186. Dong Z, Li W, Liu J et al. The rice phosphate transporter protein OsPT8 regulates disease resistance and plant growth. Sci Rep. 2019; 9: 1–10.
187. Goff SA, Ricke D, Lan T-H et al. A draft sequence of the rice genome (Oryza sativa L. ssp. japonica). Science. 2002;296:92–100.

188. Seo H-M, Jung Y, Song S et al. Characterization of a phosphate transporter gene from Arabidopsis thaliana. Plant Physiol. 1999;119:1077–87.

189. Meissner R, Michael AJ. Isolation and characterization of a phosphate transporter gene from Arabidopsis thaliana. PLoS Genet. 2010;6:e1001218.

190. Delhaize E, Randall PJ. Characterization of a phosphate transporter gene from Arabidopsis thaliana. Plant Physiol. 1995;107:207–13.

191. Aung K, Lin S-I, Wu C-C et al. Molecular characterization of a novel phosphate transporter gene from Arabidopsis thaliana. Planta. 1998;205:251–6.

192. He I, Zhao M, Wang Y et al. Phloem transport and functional diversification of the plant PHOSPHATE1 gene family: a focus on Glycine max. BMC Evol Biol. 2013;13:103–13.

193. Salazar-Vidal MN, Acosta-Segovia E, Sanchez-Leon N et al. Characterization and transposon mutagenesis of the maize (Zea mays) Pho1 gene family. PLoS One. 2016;11:e0161882.

194. Secco D, Baumann A, Poirier Y. Characterization of the rice PHO1 gene family reveals a role for OsPHO1;2 in phosphate homeostasis and the evolution of a distinct clade in dicotyledons. Plant Physiol. 2010;152:1693–704.

195. Gonzalez E, Solano R, Rubio V et al. PHOSPHATE TRANSPORTER TRAFFIC FACILITATOR1 is a plant-specific SEC12-related protein that enables the endoplasmic reticulum exit of a high-affinity phosphate transporter in Arabidopsis. Plant Cell. 2005;17:3500–12.

196. Devaiah BN, Karthikeyan AS, Raghothama KG. WRKY75 transcription factor is a modulator of phosphate acquisition and root development in Arabidopsis. Plant Physiol. 2007;143:1789–801.

197. Meissner R, Michael AJ. Isolation and characterization of a diverse family of Arabidopsis thaliana and three-fingered C2H2 zinc finger protein genes and cDNAs. Plant Mol Biol. 1997;33:615–24.

198. Chen Y-F, Li L-Q, Xu Q et al. The WRKY6 transcription factor modulates PHOSPHATE1 expression in response to low pi stress in Arabidopsis. Plant Cell. 2009;21:3554–66.

199. Rubio G, Walk TC, Ge Z et al. Root gravitropism and below-ground competition among neighbouring plants: a modelling approach. Ann Bot. 2001;88:929–40.

200. Miura K, Rus A, Shakhruh A et al. The Arabidopsis SUMO E3 ligase SIZ1 controls phosphate deficiency responses. Proc Natl Acad Sci. 2005;102:7760–5.

201. Miura K, Lee J, Gong Q et al. SIZ1 regulation of phosphate starvation-induced root architecture remodeling involves the control of auxin accumulation. Plant Physiol. 2011;155:1000–12.

202. Duan H-Y, Shi L, Ye X-S et al. Identification of phosphorus efficient germplasm in oilseed rape. J Plant Nutr. 2009;32:1148–63.

203. Duan H-Y, Shi L, Ye X-S et al. Constitutive expression of phosphate transporter, OsPnt1;1, modulates phosphate uptake and translocation in phosphate-replete rice. Plant Physiol. 2012;159:1571–81.

204. Ostrowski M, Rus A, Shakhruh A et al. Identification of phosphorous efficient germplasm in oilseed rape. Horticulture Research. 2022;9:uhac011

205. Dong B, Rengel Z, Delhaize E. Uptake and translocation of phosphate by pho2 mutant and wild-type seedlings of Arabidopsis thaliana. Plant Physiol. 2007;143:1112–22.

206. Lv Q, Zhong Y, Wang Y et al. SPX4 negatively regulates phosphate signaling and homeostasis through its interaction with PHR2 in rice. Plant Cell. 2014;26:1586–97.

207. Wang Z, Ruan W, Shi J et al. Rice SPX1 and SPX2 inhibit phosphate starvation responses through interacting with PHR2 in a phosphate-dependent manner. Proc Natl Acad Sci. 2014;111:14953–8.

208. Smith AP, Jain A, Deal RB et al. Histone H2A.Z regulates the expression of several classes of phosphate starvation response genes but not as a transcriptional activator. Plant Physiol. 2010;152:217–25.

209. Chen Z-H, Nimmo GA, Jenkins GI et al. BHLH32 regulates several biochemical and morphological processes that respond to phosphate starvation in Arabidopsis. Biochem J. 2007;405:191–8.

210. Ramaiah M, Jain A, Raghothama KG. Ethylene response Factor070 regulates root development and phosphate starvation-mediated responses. Plant Physiol. 2014;164:1484–98.

211. Devaiah BN, Madhuvanthi R, Karthikeyan AS et al. Phosphate starvation responses and gibberellic acid biosynthesis are regulated by the MIR62 transcription factor in Arabidopsis. Mol Plant. 2009;2:43–58.

212. Shin H, Shin H-S, Chen R, Harrison MJ. Loss of At4 function impacts distribution between the roots and the shoots during phosphate starvation. Plant J. 2006;45:712–26.

213. Shen C, Wang S, Zhang S et al. OsARF6, a transcription factor, is required for auxin and phosphate starvation response in rice (Oryza sativa L.). Plant Cell Environ. 2013;36:607–20.

214. Wang S, Zhang S, Sun CD et al. Auxin response factor (OsARF12), a novel regulator for phosphate homeostasis in rice (Oryza sativa). New Phytol. 2014;201:91–103.

215. Hurley BA, Tran HT, Marty NJ et al. The dual-targeted purple acid phosphatase isozyme AtPAP26 is essential for efficient acclimation of Arabidopsis to nutritional phosphate deprivation. Plant Physiol. 2010;153:1112–22.

216. Okazaki Y, Otsuki H, Narisawa T et al. A new class of plant lipid is essential for protection against phosphoryl stress. Nat Commun. 2013;4:1–10.

217. EURISCO. EURISCO Catalogue. http://eurisco.ecpgr.org (2020).

218. Payne AC. Harnessing the genetic diversity of watercress (Rorippa nasturtium-aquaticum) for improved morphology and anti-cancer benefits: underpinning data for molecular breeding. University of Southampton. 2011.

219. USDA. Germplasm Resources Information Network. https://npgsweb.ars-grin.gov/gringlobal/search (2020).

220. Voutsina, N. Elucidating the genomics of nutritional and morphological traits in watercress (Rorippa nasturtium-aquaticum) for improved morphology and anti-cancer benefits: underpinning data for molecular breeding. University of Southampton. 2011.

221. Di Noia J. Defining powerhouse fruits and vegetables: a nutrient density approach. Preu Chronis Dis. 2014;11:e95.

222. Poikane S, Kelly MG, Herrero FS et al. Nutrient criteria for surface waters under the European water framework directive: current state-of-the-art, challenges and future outlook. Sci Total Environ. 2019;695:133888–14.