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Research Article

Modeling reveals synergies among root traits for phosphorus acquisition in pearl millet

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ABSTRACT

Pearl millet is a key food security grain crop in the world's drylands due to its tolerance to abiotic stresses. However, its yield remains low and is negatively impacted by climate change. Root phenes are potential targets to improve crop productivity and resilience to environmental stress. However, the sheer number of combinations resulting from interactions of multiple phenes is a challenge for empirical research. *In silico* approaches are a plausible alternative to assess the utility of different phene combinations in varying states over diverse environmental contexts. Here, we developed an implementation of the functional-structural plant/soil model – OpenSimRoot, for pearl millet in typical sub-Sahelian soil and environmental conditions. Root architectural, anatomical, and physiological parameters were measured using a popular pearl millet variety (Souna 3) and implemented in the model. The above-ground biomass and root length density predicted by the model were similar to data from field trials. The utility of different root phenes was then evaluated for improved phosphorus uptake and plant growth in P deficient soils. Doubled root hair length and density, shallower root angle (−15°) and doubled long lateral root density were found to improve plant growth by 76 %, 33 % and 33 % respectively under low P conditions. Moreover, these phenes showed synergism when combined *in silico* and led to optimal biomass production in low P supply conditions that resulted in a 75 % loss of biomass in the reference variety. Our study suggests that these phenotypes could be targeted to improve biomass production in pearl millet and consequently its yield in low-P availability conditions.

1. Introduction

Pearl millet (*Pennisetum glaucum* (L.) R. Br.) is the sixth most important cereal crop in terms of global production and one of the most drought and low soil fertility tolerant crops [1–3]. It is cultivated on ~27 million hectares worldwide in regions where annual rainfall is between 200 and 500 mm and it is the staple crop for more than 90 million people [4,5]. Compared to other cereal crops, pearl millet is a rich source of nutrients such as proteins, lipids, calcium, phosphorus, essential amino acids, iron and zinc [6]. It is, therefore, a very important crop for food and nutritional security in arid and semi-arid regions of sub-Saharan Africa and India [3].

In West Africa, pearl millet is mainly grown by smallholders in sandy soils and in low input agrosystems [7,8]. Phosphorus (P) availability is a major constraint to pearl millet productivity in many parts of West Africa as P availability in sandy soils of the Sahel is often below 2 mg kg^{-1} [9,10]. Furthermore, P is highly stratified in the soil and mostly found in superficial layers [11]. Therefore, P acquisition is highly dependent on

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the architecture of the root system [12].

Root system architecture, the spatiotemporal configuration of a root system, determines the ability of a plant to exploit soil resources [11, 13–15]. The pearl millet root system is characterized by a fast-growing primary root that rapidly colonizes deep soil horizons and by crown and lateral roots that begin to emerge 6 days after germination [16,17]. It can reach up to 3 m in depth and can exploit soil volumes of up to 6 m^3 at low planting densities due to its lateral propagation [18]. Three types of lateral roots were identified in pearl millet, differing in growth phase, length, diameter, and anatomy [16,19]. Under water stress, pearl millet grows deeper roots by favoring axial root elongation and reducing root length density in superficial soil horizons [20,21]. These characteristics may enhance its ability to access water present in deep soil horizons but impair P capture from relatively enriched superficial soil horizons.

Phenes are the basic unit of the phenotype like genes are basic components of the genotype [22,23]. Root phenes that improve exploration of the topsoil have been shown to improve P acquisition and have been used to create new crop varieties with improved yield in P limited low input agrosystems in Africa [8,12,22]. These include root angle [24, 25], the number of basal roots [26,27] or root hair length and density [28–33]. Root phenes interact with each other to determine the adaptive value of plants, this interaction can be either synergistic, additive, or antagonistic [34]. The evaluation of these interactions considers the interaction of the different states of the phenes and their interactions with the environment, leading to a large number of scenarios [35]. The use of *in silico* models has been instrumental to explore the large number of root phene combinations that might improve performance under low P conditions [23,24].

In this study, we developed a functional-structural root model for pearl millet using the OpenSimRoot framework. OpenSimRoot is a feature-rich and widely published functional-structural plant and soil model [36–38]. It has been developed for several plants such as maize, squash, bean, lupin, *Arabidopsis,* barley and rice [31,35,39–46]. It is Open Source and maintained by an international community of root researchers who continuously improve the model. Compared to other well-known models such as RootTyp [47], R-SWMS [48] or RootBoox [49], OpenSimRoot can simulate root growth, water and nutrient dynamics in 3D soil and shoot processes in non-structural manner. OpenSimRoot is an effective tool for analyzing root geometry and various root traits across different climates, nutrient and water regimes, and soil types, thus informing the development of optimal root ideotypes for nitrogen, phosphorus, and potassium uptake, as well as resilience to drought and compacted soil. This heuristic plant model was coupled to a soil model that was parameterized with typical Sahelian sandy soil characteristics, allowing the simulation of pearl millet root systems in realistic environments. It was then used to analyze the impact of changes in root architectural and anatomical phenes as well as their potential synergies on P acquisition and plant growth.

2. Material and methods

2.1. Model description

A 3D functional-structural plant/soil model, OpenSimRoot, which combines a three-dimensional representation of root systems with physiological function models [37,50] was used to simulate root growth, root-soil interactions, soil resource capture and shoot growth of a pearl millet plant under conditions of variable soil phosphorus availability. OpenSimRoot is an open-source, extended version of SimRoot [36], that has a unique conception based on the coupling of different mini-models, its code is modular and allows the construction of different models based on an XML meta-language [37]. OpenSimRoot allows simulation of root systems based on growth, branching and functioning rules for different classes of roots composed of small discrete segments (~1 cm) linked together, leading to a complete root system architecture. With a three-dimensional soil component, the model dynamically simulates root growth and soil resource acquisition. It models photosynthesis, carbon allocation, nutrient and water fluxes in the soil, nutrient and water uptake by roots, responses to nutrient stress, root responses to local nutrient conditions and many other characteristics important for plant development [37].

2.2. Plant material and growth conditions

The pearl millet variety Souna3 was used in this study. It is a dual-purpose pearl millet variety selected for both fodder and grain with a short cycle of around 90 days that is widely cultivated in West Africa. Model parametrization trials were conducted at CERAAS (Senegal; 14°79 N, 16°89W) during the dry season 2020 (October to December), 2021 (April to June), and 2022 (March to May). Plants were grown in a nethouse in a sandy soil representative of the pearl millet growing areas in West Africa. This soil is a weakly leached tropical ferruginous soil [51], rich in sand (95 % in total) with a clay fraction dominated by kaolinite, a pH between 5.4 and 6.1, and low water retention capacity [52]. Before sowing, a light plowing was carried out once using a disc plow and fertilization was applied to a rate of 150 kg. ha^{-1} of 15N–10P–10K, as recommended for pearl millet. Plots consisted of 5 lines containing 20 hills per line, with 80 cm between lines and 80 cm between hills within the lines, as in farmers' fields. After two weeks, hills were thinned to one plant and additional fertilization with 50 kg. ha^{-1} of 15N–10P–10K was applied. The plants received 30 mm of water twice a week until harvest.

2.3. Phenotyping

In each trial, three plants were harvested twice a week for root and shoot phenes measurements from 6 to 35 days after sowing. For each plant, the number of tillers was counted, and the leaf area was measured using a planimeter (Licor-3600c). Shoot fresh weight was measured as well as shoot dry weight after two days in an oven at 60 °C. Roots were harvested using the shovelomics method [53] and washed to remove soil particles. The root angle at each node, the number of roots per node and the distance between nodes were measured. Two representative roots segments of 2 cm were sampled at each node at around 1 cm from the base of the stem and preserved in 70 % (v/v) ethanol. Roots were embedded in agar 2 % and transversal sections were produced using a vibratome (Leica VT1000 S) and imaged using a microscope (Olympus Bx50) equipped with a digital camera (Micro Publisher 3.3 RTV). The diameter and number of xylem vessels were measured using the ImageJ software (http://www.mecourse.com/landinig/software/software.html). A subset of root samples was imaged using laser ablation tomography [54]. Samples were critically-point dried (Leica CPD300), ablated using a q-switched, pulsed UV laser at 355 nm (Edgewave Model PX100-3-GF) and cross-sections imaged using a 5× infinity-corrected long working distance objective (Mitutoyo) connected to a machine vision camera (FLIR Grasshopper 3). Images were processed for anatomical trait measurements using a modified version of RootScan software [55]. Photosynthesis was measured using a CI 340 probe (CID Bio-Science) on the last fully expanded leaf of three plants twice a week on sunny days between 10 and 12 a.m. Root respiration was measured with the Licor 6200 (LI-COR Biosciences).

2.4. Model parametrization

The measured plant phenes used to parameterize a pearl millet OpenSimRoot model are listed in Table 1.

Environmental parameters, i.e. climate and edaphic factors, were parameterized using climatic and soil data collected from the ISRA research station of Bambey (Centre National de Recherches Agronomiques de Bambey) in Senegal (from Khouma [52]). The Rosetta3 software [57] was used to estimate van Genuchten's water retention parameters to accurately capture the soil hydraulic in the model [58]. Precipitation was parameterized in the model at 30 mm twice a week.

Table 1

Phenotypic data used for model parameterization.

Phenotypic data from trials	Phenotypic data from literature
root angle	lateral root density [19]
number of nodes	lateral root diameters [16]
number of roots per node	lateral root growth rate [16]
distance between nodes	albedo, latitude, altitude [56]
time of appearance of nodes	bulk density (soigrid.org)
root diameter	
root hair length	
root hair density	
number of xylem per root type	
tiller number	
root growth rate	
leaf area	
Photosynthesis	
root respiration	

2.5. Root impact calculation from simulations

Root growth of six Souna3 plants was simulated for 60 days, and output of the model was compared with empirical field measurements of root length density from Faye et al. [21]. Simulations were repeated three times. Virtual trenches were then made with paraview [59] at 10 and 30 cm from the middle plant within the virtual row (as performed *in vivo* in Faye et al. [21]). The number of roots crossing the virtual trench, i.e the number of root impacts, were counted using a grid of 20 × 20 cm within the soil profile. Root impacts counted along the soil depth in the different simulations were compared with those measured in the field [21].

2.6. Simulated scenarios

Simulations were performed on a single plant using the planting density usually applied in farmers' fields (0.8 m distance between and within row) within a soil depth of 3 m.

Details of all the virtual experiments are described in Table 2.

2.7. Evaluation of phene interactions

Interactions among phenes were quantified as previously described [31]. In summary, to assess the types of interaction for each phene (synergism, antagonism or additivity), the effects of each individual phene state were calculated by subtracting the simulated shoot biomass for control conditions under low P (P inducing 75 % biomass loss: 6.8 kg. ha^{-1}), from that of each individual phene state. This difference is referred to here as the delta and represents the quantitative effect of a change of phene state on biomass production. These differences (delta) were then added to the value of the control phenotype according to the type of phene combination to estimate the expected additive effect of the phene states. If the biomass gain of a simulated plant combining different phenes was greater than the sum of the gain for each phene state, the phenes were considered as synergistic. If the gain was less than the sum of gains, then the phenes were considered antagonistic. If the gain was similar to the sum of gains, the phenes were considered additive.

Table 2

Root phenes combinations and experimental design used in the simulations of pearl millet root architecture.

Target root phenes or environmental condition	States	Units	No. of states
Root hair density	x0.25, x0.5, x2, x4	#.cm^{-2}	4
Root hair length	x0.25, x0.5, x2, x4	cm	4
Nodal root angle	−15, +15	Degree	2
Lateral root branching density			
Lateral root type A	x0.5, x2	#.cm^{-1}	2
Lateral root type B	x0.5, x2	#.cm^{-1}	2
Lateral root type C	x0.5, x2	#.cm^{-1}	2
Soil phosphorus concentration	6.5, 7.8, 8.7, 10.4	kg.ha^{-1}	4

2.8. Statistical analyses

R software (version 4.2) was used for statistical analyses [60]. A one-factor ANOVA (*anova.lm* function) was carried out to compare the root impacts measured in the field with the root impacts measured on the models for each soil horizon. The parameters of the model output table were processed with Excel and plotted with R.

3. Results

3.1. Development of a pearl millet OpenSimRoot model

To identify root phenes that could contribute to better yield of pearl millet in low soil phosphorus conditions, a functional structural model of the pearl millet root system was created using the OpenSimRoot framework based on parameters measured on the short cycle Souna3 variety (see Table 1 for the list of parameters implemented in the model). The model simulates the 3 types of lateral root reported in pearl millet (Fig. 1A and B). The shoot dry weight predicted by the OpenSimRoot pearl millet model was comparable with shoot dry weight measured for Souna3 in field conditions by Faye and al [21]. and in our parameterization trials (Fig. 1C). We compared the root architecture outputs from the model with root length density derived from root impacts data obtained on Souna3 in field conditions [21]. To do so, a 6-plants plot was simulated (Fig. 1D) and a virtual trench was created between the lines 10 or 30 cm away from a plant and root impacts from the model were measured on this virtual trench to mimic the experimental setup used in Faye and al [21]. Three independent simulations were run, and simulated values were compared to the root impacts measured in field conditions [21]. No significant differences were observed between simulated and measured root impacts, except at a depth of 40 cm at 10 cm from the plant (*p*-value <0.000767; Fig. 1E) and below 120 cm depth at both a distance of 10 and 30 cm from the plant (*p*-value <0.00163 and *p*-value $<3.39.10^{-5}$ respectively; Fig. 1F). In both cases, the simulated values were higher than the measured values. For deep roots (>120 cm), this discrepancy is most likely due to the underestimation of root length density at depth in the field due to the experimental methodology (see Faye and al. [21]). Altogether, we conclude that the simulated output of the model in terms of shoot biomass production and root length density was comparable with data measured in field experiments.

3.2. Evaluation of the impact of individual root phenes on phosphorus acquisition

To evaluate the effect of P deficiency on plant growth, we first performed simulations at different soil P availability levels (SupFig. 1). We observed a sharp decline in simulated biomass for P soil availability level less than 135 kg ha^{-1} which corresponds to 9.8 kg.ha-^{1} of P available in the soil. This value was therefore used as the optimal value in our simulations. The data were then used to identify the soil P availability levels leading to 25 %, 50 % and 75 % biomass reduction *in silico* (corresponding to 6.8 kg.ha-^{1}, 7.7 kg.ha-^{1} and 8.5 kg.ha-^{1} of P available in the soil, respectively). These P availability levels were chosen to cover a wide range of low-P availability levels ranging from mild to strong P deficiency. They were subsequently used to test the impact of root phenes on P acquisition and plant development in conditions of P deficiency in our simulations.

Root hairs are important for phosphorus acquisition [12,61]. We therefore first tested the impact of changes in root hair length and density on phosphorus acquisition and plant growth in conditions of phosphorus deficiency. Increased root hair length (Fig. 2A, SupFig. 2) or density (Fig. 2B–SupFig. 2) was associated with improved plant growth and P

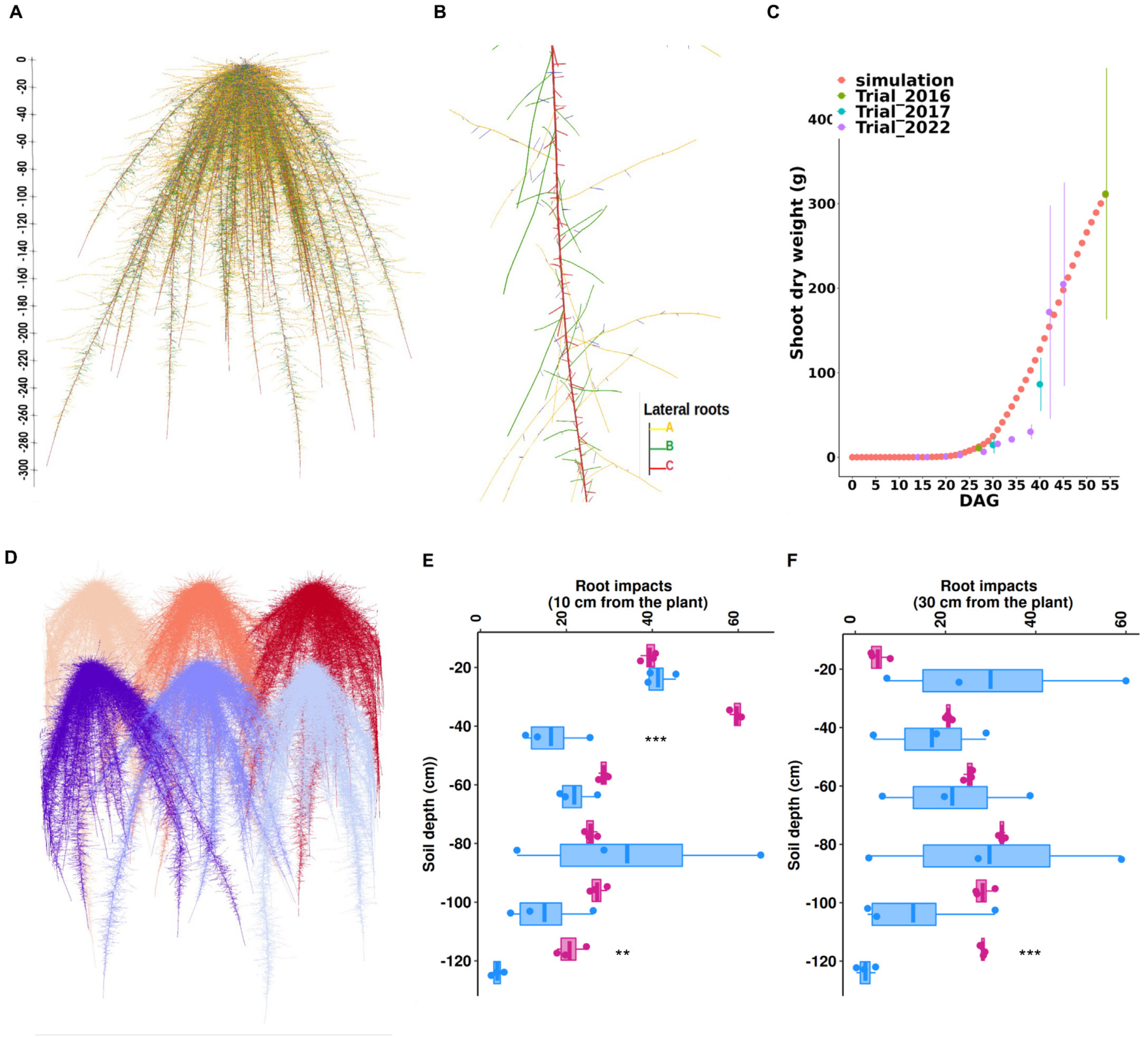

Fig. 1. Outputs of the OpenSimRoot pearl millet model. (A) 3D representation of a simulated root system in optimal P conditions 43 days after sowing. (B) Detail of a simulated root system showing the different types of lateral roots. (C) Comparison plant biomass simulated by OpenSimRoot pearl millet and measured in different field trials under optimal P conditions. DAG: days after germination. (D) Predicted and measured root impacts on a plane 10 cm from the plant. (E) Predicted and measured root impacts on a plane 30 cm from the plant.

acquisition in all soil P availability levels. On the contrary, reducing root hair length or density impaired plant growth (Fig. 2A and B). Increasing root hair density had a stronger positive impact on plant growth under P deficiency than increasing root hair length (Fig. 2A and B; Table 3). Combining the two phenes improved biomass production at all P availability levels (Fig. 2C). When we quantified the interactions between root hair density and root hair length for shoot biomass under low P availability levels, we found that combining increased root hair length and density had a synergistic effect (Table 3, Fig. 2C). Altogether, this suggests that longer and denser root hairs could lead to improved tolerance to low-phosphorus conditions in pearl millet.

As phosphorus is mainly present in the topsoil, shallow root systems have also been associated with better P acquisition [11,62,63]. We therefore simulated the impact of changes in crown root angles in pearl millet. We observed that increasing root angle by 15° relative to the soil surface (i.e.e deeper root systems, Fig. 3A) led to slightly decreased biomass production for all phosphorus levels (Fig. 3B). On the other hand, decreasing root angle by 15° (i.e. a shallower root system) increased biomass production in low phosphorus conditions (Fig. 3B). Thus, decreasing crown root angle could improve phosphorus acquisition in pearl millet.

Previous root phenotyping experiments revealed that pearl millet develops 3 types of lateral roots with different anatomies and growth patterns [16,19]. Type A laterals have an anatomy similar to the primary root, an indeterminate root growth and can form secondary laterals. Type B laterals are thinner than type A, have less cell layers and show a determinate root growth. Finally, type C laterals are very thin (68–140 µm diameter) with only two or three xylem vessels, no aerenchyma, no branching and a short, determinate, growth period [16,19]. In previous experiments, type A, B and C laterals were shown to represent 13 %, 24 %

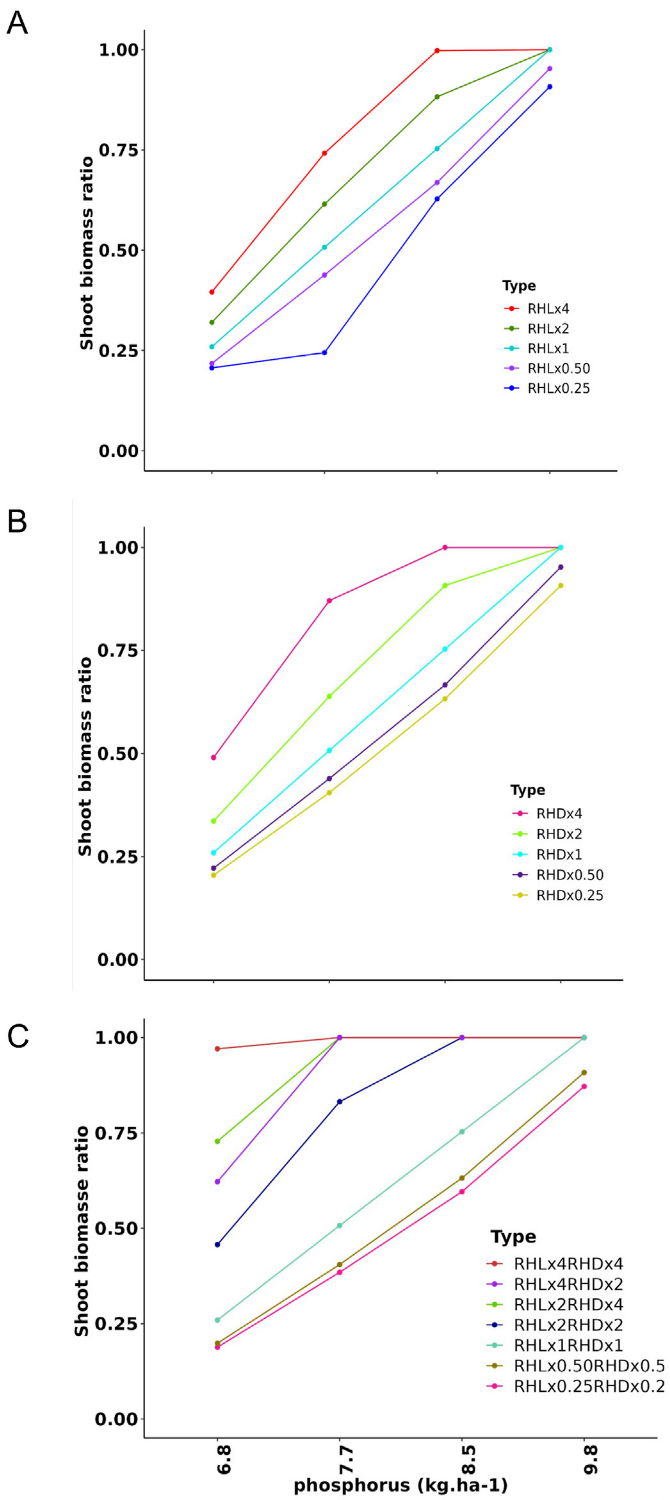

Fig. 2. Influence of root hair length and density on tolerance to P deficiency in pearl millet. (A) Impact of changes in root hair length on the shoot biomass ratio (expressed relative to biomass production by the control plant in optimal conditions) in different P conditions. (B) Impact of changes in root hair density on the shoot biomass ratio in different P conditions. (C) Synergism between root hair length and density on shoot biomass ratio in different P conditions.

and 63 % of the total laterals respectively [19]. These different lateral root types were included in the model. To test their importance for P acquisition, we modeled the impact of changes in their density on plant growth in P-deficient conditions. First, we simulated pearl millet plants having only one type of lateral root. In all cases, these *in silico* phenotypes performed worse than control plants (Souna3) for all soil P availability levels (Fig. 4A). However, plants having only type A laterals were less affected than the other phenotypes (Fig. 4A). Similarly, simulated plants lacking one type of lateral root but retaining the other 2 types had reduced biomass production for all soil P availability levels (Fig. 4B). Interestingly, phenotypes that retained type A lateral roots performed better than the other phenotypes in all phosphorus availability levels. Hence, these simulations suggest that type A lateral roots are more important for P capture than the other root types. However, this approach, analogous to a loss-of-function, led to overall reduced lateral root density by the removal of some lateral root types from the model.

Next, to avoid the impact of reduced lateral root density, we created *in silico* phenotypes that could make just one type of lateral root but at a similar total lateral root density than in the control model i.e. replacing all lateral roots by one lateral root type. We observed that plants that formed only A or B laterals outperformed control plants for all low P availability levels, with plants having A types performing best (Fig. 4C). On the other hand, plants having only C-type lateral roots performed poorly for all soil P availability levels (Fig. 4).

Finally, we kept all 3 lateral root types in the model and changed the density of one type of lateral while keeping the other unchanged. This mimics breeding for increased density of one type of lateral root. Increasing the density of all types of lateral roots led to increased plant growth and phosphorus acquisition in low-P soil compared to the control condition (Fig. 4D). Consistent with previous simulations, increasing type A lateral root density gave the best results in all low-P conditions (Fig. 4D). Altogether, this indicates that having a root system with more long laterals is beneficial for P capture and biomass production in conditions of suboptimal P availability.

Aerenchyma formation was associated with reduced metabolic cost of root growth and therefore to more developed root systems [62]. We therefore tested the impact of changes in the proportion of aerenchyma formation on plant growth in low-P conditions. Increasing or decreasing aerenchyma formation has a very limited effect on biomass production in low P conditions (SupFig. 4). This suggests that aerenchyma formation is not a target trait to improve tolerance to P-deficiency in pearl millet.

3.3. *Evaluation of phene synergisms*

In order to test how the phene states that we found to have a positive impact on low-P tolerance *in silico* could be combined, we next quantified how they interact with each other. Simulations were performed using different combinations of phene states that had the optimal positive impact on plant growth under low-P conditions (increased root hair density and length, decreased crown root angle and increased density of type A lateral roots) at a low-P availability level corresponding to a biomass loss of 75 % for the control model. When comparing the impact of individual phene states, increasing root hair density and length had the strongest positive effect on biomass production (Table 3). A synergistic effect was found when combining root hair phenotypes with either crown root angle or increased type A lateral root density or the 3 phenotypes together (Table 4).

Finally, we simulated a virtual ideotype combining the 3 phenotypes and compared it to Souna3 (control) at optimal and low P availability levels (Fig. 5A and B). This ideotype outperformed the reference variety (Souna3) and was able to maintain optimal biomass production (Fig. 5C) and improved P uptake (Fig. 5D) at low P availability levels. We conclude that synergism between root hair length and density, crown root angle and type A lateral root density could be exploited to create varieties more adapted to low P soils.

4. Discussion

Pearl millet is an important cereal crop for food security in arid and

Table 3

Synergistic effect of root hair length and density under low P availability. Simulations were performed for P availability leading to a 75 % loss in biomass production in the control (Souna3). Simulated biomass: biomass output from the OpenSimRoot model for the target phenotype; Delta: difference between the simulated biomass for a phenotype and the simulated biomass for the control (Souma3); Expected biomass if additive affect: sum of the control phenotype and the delta for each individual phene state that were associated; Additive effect: sum of the delta for each individual phene state that were associated; Observed effect: Difference between the simulated biomass for a given phenotype and the biomass of the control plant; Observed effect over additive effect: Ratio between the observed effect and the additive effect, if negative, the traits are antagonistic, if negative, the traits are synergistic.

Phenotype	Simulated biomass (g)	Delta (g)	Expected biomass (g) if additive effect	Additive effect (g)	Observed effect (g)	Observed effect over additive effect
Control	40.02	–	–	–	–	–
RHLx0.25	31.91	−8.11	–	–	–	–
RHLx0.50	33.58	−6.43	–	–	–	–
RHLx2	49.39	9.37	–	–	–	–
RHLx4	61.07	21.05	–	–	–	–
RHDx0.25	31.58	−8.44	–	–	–	–
RHDx0.50	34.21	−5.81	–	–	–	–
RHDx2	51.84	11.82	–	–	–	–
RHDx4	7565	35.64	–	–	–	–
RHLx2RHDx2	70.56	–	61.21	+21.197	+30.55	+44.11 %
RHLx4RHDx4	149.77	–	106.08	+56.688	+100.39	+77.08 %
RHLx2RHDx4	112.32	–	106.08	+45.011	+62.93	+39.82 %
RHLx4RHDx2	95,98	–	93,94	+32,874	+55,97	+70,25 %

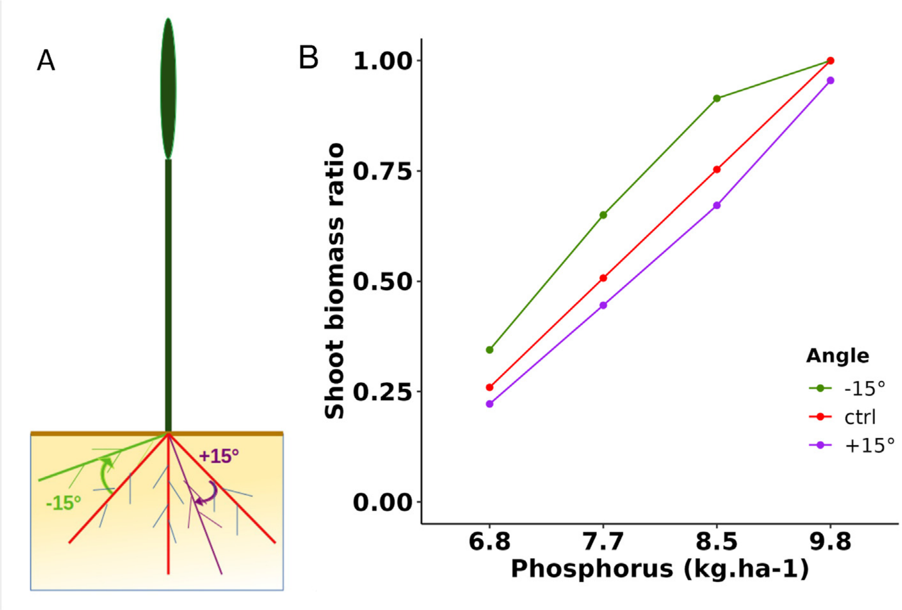

Fig. 3. Influence of crown root angle on tolerance to P deficiency in pearl millet. (A) Schematics showing the modifications of root angle performed in OpenSimRoot pearl millet. (B) Impact of changes in crown root angle on shoot biomass ratio in different P conditions.

semi-arid regions of Africa and India with low rainfall and poor soil fertility. Suboptimal phosphorus availability is one of the main factors limiting its yield in sub-Saharan Africa. We previously showed that pearl millet roots have a low level of colonization by arbuscular mycorrhizal fungi and a low mycorrhizal dependency and therefore relies more on its root system for hydromineral nutrition [64]. Here, we implemented the functional-structural modeling framework OpenSimRoot for pearl millet to explore root phenotypes that could be targeted to improve P acquisition and plant production in low-P environments. As observed in other cereals [34,65], our simulations suggested that increasing root hair density and length and decreasing crown root angle relative to the soil surface improve P acquisition by enhancing topsoil exploration. As reported for maize, the model predicted synergism between root hair length and density for biomass production under low P availability [31]. On the other hand, the model did not predict any significant change in biomass production or P acquisition when increasing aerenchyma formation. This is different from what was reported in maize [27,66,67] and might be explained by the lower root diameter and respiration rate in pearl millet compared to maize that might limit the metabolic cost of root growth in the former compared to the latter. As pearl millet develops different types of lateral roots, we also studied the impact of the different lateral root types on P uptake and plant growth. We found that increasing the density of long, indeterminate type A lateral roots was beneficial for phosphorus acquisition. Interestingly, we found that combining denser and longer root hairs, shallower crown root angle and higher density of long type A lateral root had a very strong synergistic effect. This indicates that breeding for an integrated root phenotype combining these phene states could lead to a strongly improved P acquisition and plant growth in low-P agrosystems.

Among potential target phenes, root hairs are promising targets because they have a strong impact on phosphorus acquisition at very low metabolic cost, in addition to their positive effect on the uptake of other ions [61]. Therefore, there is no expected trade-off for increased root hair length and density. Root hair phenes are easy to phenotype at high throughput in controlled conditions [33,68]. Phenotypic diversity for root hair length is available in pearl millet [17] and QTLs for root hair traits have been described in other cereals [68,69]. Altogether, this suggests that breeding for increased root hair length and/or density is

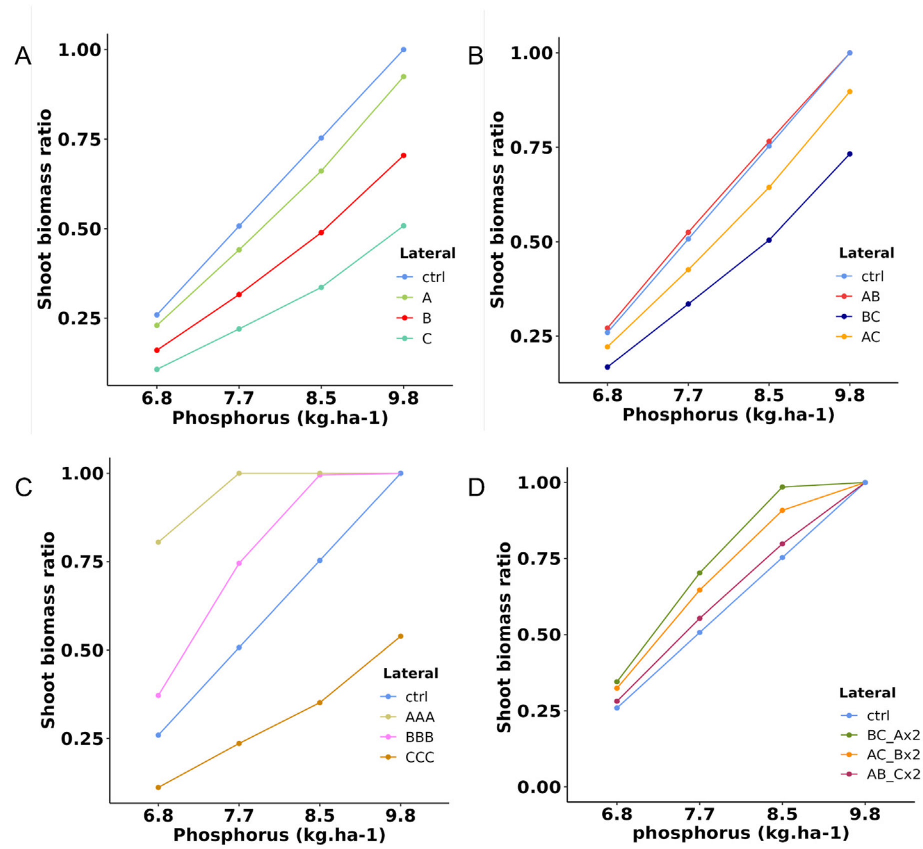

Fig. 4. Influence of the different types of lateral roots on tolerance to P deficiency in pearl millet. (A) Impact of the loss of 2 lateral root types on shoot biomass ratio in different P conditions. (B) Impact of the loss of 1 lateral root type on shoot biomass ratio in different P conditions. (C) Impact of changing all lateral roots to one type on shoot biomass ratio in different P conditions. (D) Impact of changing the density of one type of lateral root on shoot biomass in different P conditions.

Table 4

Synergism of root hair length, root hair density, lateral root type B and nodal root angle under suboptimal P availability. Simulations were performed for P availability leading to a 75 % loss in biomass production in the control. Simulated biomass: biomass output from the OpenSimRoot model for the target phenotype; Delta: difference between the simulated biomass for a phenotype and the simulated biomass for the control (Souma3); Expected biomass if additive affect: sum of the control phenotype and the delta for each individual phene state that were associated; Additive effect: sum of the delta for each individual phene state that were associated; Observed effect: Difference between the simulated biomass for a given phenotype and the biomass of the control plant; Observed effect over additive effect: Ratio between the observed effect and the additive effect, if negative, the traits are antagonistic, if negative, the traits are synergistic.

Phenotype	Simulated biomass (g)	Delta (g)	Expected biomass (g) if additive effect	Additive effect (g)	Observed effect (g)	Observed effect over additive effect
Control	40,02	–	–	–	–	–
Angle-15	53,14	13,12	–	–	–	–
lateralBC_Ax2	53,33	13,31	–	–	–	–
RHLx2RHDx2	70,56	30,55	–	–	–	–
RHLx2RHDx2 + LateralBC_Ax2	92,34	–	83,87	+43,86	+52,32	+19,30 %
RHLx2RHDx2 + Angle-15	92,26	–	83,68	+43,67	+52,24	+19,64 %
Angle-15 + LateralBC_Ax2	68,88	–	66,45	+26,43	+28,86	+9,20 %
RHLx2RHDx2 + Angle-15+ LateralBC_Ax2	154,28	–	127,54	+56,98	+114,27	+100,55 %

both feasible and would lead to many benefits for pearl millet yield in low input agrosystems.

Root angle, another key trait for P acquisition, is also easy to phenotype and QTLs have been found in other cereal crops [70,71]. However, steeper root angle is also important for root system depth and for water and mobile nutrients (such as nitrogen) acquisition for deeper soil domains. A root system with some proportion of steeper and shallower crown roots might offer a tradeoff for P, N and water uptake. The impact of altering crown root angle on water and nitrogen acquisition along with P uptake will need to be further explored in pearl millet.

Lateral root density might have more limited trade-offs. Different types of lateral roots have been reported in rice, maize and pearl millet [16,19,72] but the functional significance of this diversity is unknown. Here we found that type A, i.e. long lateral root showing an indeterminate growth pattern, are more important for phosphorus acquisition than the other two types in pearl millet and that increasing their density could improve tolerance to low P availability. This is consistent with the proposed role of these lateral roots in increasing the volume of soil explored by the root system [16]. Interestingly, a QTL for long lateral root density was recently shown to improve P uptake in rice [73]. Large diversity has already been reported for lateral root density in pearl millet [16].

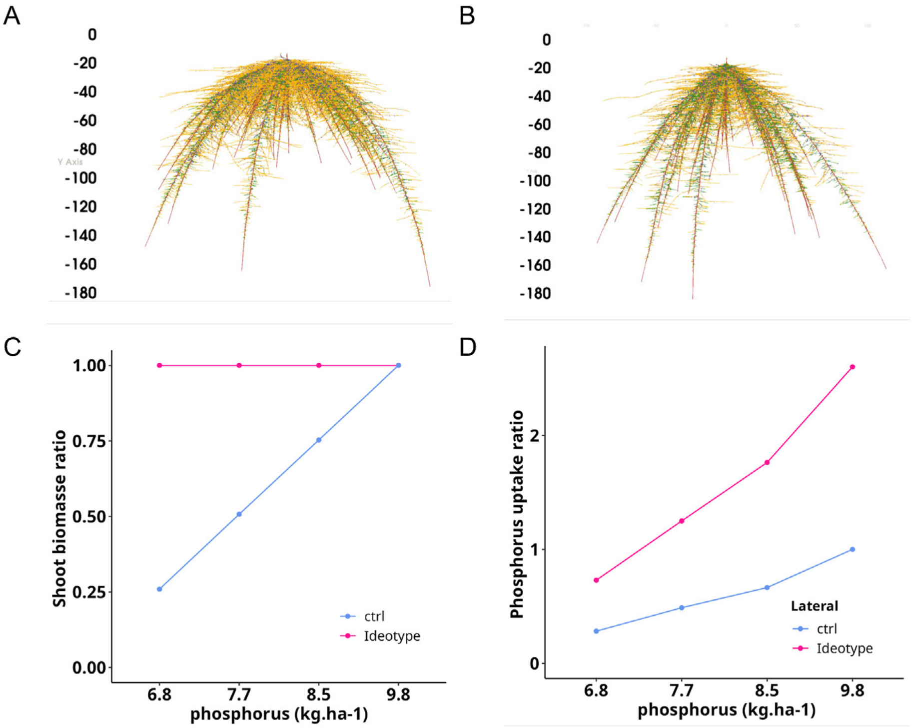

Fig. 5. An ideotype combining 3 root phenes maintains optimal biomass production and improves P uptake at low P concentration. 3D representation of the ideotype (A) and Souna3 (B) root system in low-P conditions 43 days after sowing. (C) Simulated shoot biomass ratio in the ideotype and control (Souna3) in different P conditions. (D) Simulated P uptake in the ideotype and control (Souna3) in different P conditions.

5. Conclusion

We developed an OpenSimRoot model for pearl millet and used it to identify root traits and their synergisms that could be targeted to improve growth and yield under low phosphorus (P) availability conditions, one of the main factors limiting crop production in West Africa. Our results led us to suggest an ideotype that could maintain plant growth and yield in low-P soils. Future research should focus on testing the predictions of the model. Genetic diversity for root traits has been reported in pearl millet (e.g. Refs. [16,17],) and could be used to analyze the impact of the root traits identified *in silico* on crop performance in low-P conditions in field trials. This could be used to breed varieties with modified root traits to improve pearl millet productivity in West Africa as it was done for common bean in Mozambique for example.

We analyzed the impact of changes in root traits on crop performance only in low-P conditions, while crops often face multiple stresses in natural conditions. In the semi-arid areas of West Africa where pearl millet is cultivated, drought is a major factor limiting crop yield. It will be interesting to use the pearl millet OpenSimRoot model to test the impact of the root traits we identified on crop performance under different drought stress scenarios and combined drought and low-P conditions.

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Data availability statement

The OpenSimRoot model and parameters are available at: https://forge.ird.fr/diade/opensimroot_pearlmillet/osr_pearlmillet.

CRediT authorship contribution statement

Mame Sokhatil Ndoye: Formal analysis, Investigation, Writing – original draft, Writing – review & editing. **Mikael Lucas:** Formal analysis, Supervision, Writing – review & editing. **Ishan Bipin Ajmera:** Formal analysis, Supervision, Writing – review & editing. **Bassirou Sine:** Supervision, Writing – review & editing. **Awa Faye:** Investigation, Writing – review & editing. **James Burridge:** Investigation, Writing – review & editing. **Mariama Ngom:** Investigation, Writing – review & editing. **Pascal Gantet:** Supervision, Writing – review & editing. **Darren M. Wells:** Investigation, Writing – review & editing. **Ndjido Ardo Kane:** Funding acquisition, Project administration, Writing – review & editing. **Jonathan Paul Lynch:** Conceptualization, Supervision, Writing – review & editing. **Abdala Gamby Diédhiou:** Supervision, Writing – review & editing. **Alexandre Grondin:** Conceptualization, Funding acquisition, Investigation, Supervision, Writing – review & editing. **Laurent Laplaze:** Conceptualization, Funding acquisition, Project administration, Supervision, Writing – original draft, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.cropd.2024.100059.

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