ARTICLE IN PRESS

Annals of Agricultural Science xxx (2017) xxx-xxx



Contents lists available at ScienceDirect

Annals of Agricultural Science

journal homepage:



Characterization of genotypic variability associated to the phosphorus bioavailability in peanut (*Arachis hypogaea* L.)

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ARTICLE INFO

Article history:
Received 24 January 2016
Received in revised form 17 November 2016
Accepted 31 January 2017
Available online xxxx

Keywords: Phosphorus Bioavailability Efficiency Bradyrhizobium Leguminous Peanut

ABSTRACT

In order to assess genotypic variability in some peanut genotypes depending on phosphorus availability, both effects of tri-calcium phosphate (TCP) and inoculation by <code>Bradyrhizobium</code> strain (BR) on morphological and physiological parameters were studied in five peanut genotypes (<code>Arachis hypogaea L.</code>), originated from two Algerian areas (Northern and Southern). The results obtained during the flowering stage of crop development, confirmed the positive effect of the contribution of tri-calcium phosphate (TCP) with <code>Bradyrhizobium</code> strains on the morphological characters (shoot biomass, root biomass, nodular biomass and leaf) and the physiological (nitrogenase activity, phosphorus absorption efficiency by roots (RPAE) and phosphorus use efficiency (PUE)) for the peanut genotypes cultivated in this experiment. Among five genotypes tested, it was noted that the Southern genotypes were more efficient to use TCP in the presence of <code>Bradyrhizobium</code> strain after a screening of these local genotypes, in particular, with phosphorus use efficiency (PUE) and shoot biomass production.

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Introduction

In the Mediterranean regions, agricultural production and crop yields are highly depending on the availability of water and nutrient element at the right time. Among these mineral nutrients, phosphorus limits the vegetable production in many soils. Especially in Mediterranean soils where most are calcareous soils (Drevon, 2009). The peanut is a leguminous which can amount on the symbiotic nitrogen fixation to satisfy its requirements of nitrogen. However, the low availability of phosphorus in the Mediterranean soils reduces the yields of this crop. The problem of this deficiency can be solved by the application of inorganic phosphate fertilizers (Wissuwa and Ae, 1999; Oburger et al., 2010). Nevertheless, approximately 80% of phosphate applied on the soil are quickly dis-integrated by calcium and thus become low available for the plants (Vance et al., 2003). The results obtained by various studies, show that the plants in a general way and the leguminous plants, in particular, react differently to phosphate contained in the soil according to the species and the varieties to which they belong. That could be explained by the morphological and physiological modifications of the roots having for the consequences of the modifications of the rhizosphere soil characteristics making it possible to increase the bioavailability of this element (Hinsinger, 2001; Vadez and Drevon, 2001; Alkama, 2010). In this context, the research of the peanut cultivars which can use most of this broad and insoluble pool of P proves to be necessary. Such cultivars which would behave not only better in the absence of the fertilization, but which should moreover be more efficient with phosphate fertilizers when the fixing of the phosphate fertilizers is a problem and when the insoluble phosphate form must be used (Bekele et al., 1983). Our objectives were to analyze the extent of intraspecific variability in phosphorus use efficiency for peanut genotypes under insoluble pool of P and to determine the morphological and physiological mechanisms of adaptation to low P availability. The exploitation of plant variability in the acquisition of unavailable P in soils is emerging as an alternate approach to improve productivity in low-nutrient environments. If successful, it would result in low-input and sustainable agriculture systems.

Peer review under responsibility of Faculty of Agriculture, Ain-Shams University.

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Materials and methods

The vegetable material consists of five (05) local genotypes of peanut of which three come from the North-eastern area of Algeria

http://dx.doi.org/10.1016/j.aoas.2017.01.004

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Please cite this article in press as: Kraimat, M., Bissati, S. Characterization of genotypic variability associated to the phosphorus bioavailability in peanut (*Arachis hypogaea* L.). Ann. Agric. Sci. (2017), http://dx.doi.org/10.1016/j.aoas.2017.01.004

(Berrihane, Boumalek and Tonga) and two others from the Southern area (Metlili and Sebseb), as described in Table 1. The crop was led under greenhouse, in pots of 10 kg containing a deficient phosphorus soil, whose main properties are mentioned in Table 2. The amount of $Ca_3(PO_4)_2$, $6H_2O$ provided was calculated according to the requirements of peanut in P (67 kg ha⁻¹), so 7 g of TCP was mixed with soil for each pot. Before being sown, the seeds were weighed (Table 1) and inoculated by rhizobial suspension $(10^9 \text{ cell ml}^{-1})$ using the sterile peat and gum arabic as adhesive (Suarez-Vasquez, 1975). The assay was periodically irrigated twice a week by an equal amount of water in which the characteristics are given in Table 3.

At flowering (56 days after planting), a leaf of the same vegetative row was photographed using a numerical camera for each plant, by taking a photo at a known distance. The leaf area is then calculated after analysis of the scanned image using «ImageJ» program (Reinking, 2007). Acetylene reducing activity (ARA) was measured with an *in-situ* method (Balandreau and Dommergues, 1970), in which 10 ml of acetylene (C_2H_2) was injected into the pot. After incubation (30 min), 5 ml of gaseous mixture was taken and acetylene produced was determined using a gas chromatograph with a flam ionization detector and a Spherosil XOB 075 10% Na₃PO₄ column.

Biomass harvested at 56 days after planting and samples of plant are washed with distilled water. Roots and stems were separated and dried at 80 $^{\circ}$ C during 36 h. After drying, biomasses are weighed and the yield per plant was taken at the end of the experiment.

Phosphorus contained in plant was digested by HNO_3 , H_2SO_4 and $HClO_4$ (10:1:4 v/v/v) and re-evaporated. The P concentration in digests was estimated at 460 nm using molybdenum ascorbic acid method (Murphy and Riley, 1962).

Two parameters are used to evaluate the phosphorus efficiency in peanut populations tested. The phosphorus absorption efficiency by roots (RPAE), reflecting the capacity of the root to absorb phosphorus soil and the phosphorus use efficiency (PUE), indicating the capacity of the plant to use P absorbed. Those were also presented and calculated in this study according to the following formulae, respectively (Pan et al., 2008):

Table 1Origin and properties of cultivated peanut genotypes

RPAE = P content of the whole plant/Root dry weight

PUE = Dry weight of whole plant/P content of the whole plant

Data collected were statistically analyzed by factorial ANOVA and Newman-Keuls test, using the SAS 9.0 program at the probability level α = 0.05.

Results

Shoot, root and nodule biomass

The genotypes differed substantially in their growth in the presence of *Bradyrhizobium* strain and TCP (Table 4). The addition of TCP alone increased shoot biomass by 22.29% for the five peanut genotypes. Metlili genotype had the lowest rate of increase, estimated at 18.67%, while the highest increase, 29.03% is registered in the genotype of Boumalek. However, this response is greater in the presence of *Bradyrhizobium* strain. Also, the smallest percentage was noted in Metlili genotype, whereas the highest (122.01%) was for Tonga genotype (Table 4).

On the other hand, root dry matters were reduced by 11.62% in presence of TCP alone with a maximum rate for Metlili genotype and a minimum rate for Sebseb genotype. With inoculation, the average rate of reduction reaches 61%, where we observed the rates of 102.71% and 83.55% in Southern genotypes (Sebseb and Metlili) against 73.90%. 23.13% and 21.70% for Tonga, Boumalek and Berrihane respectively (Table 4).

TCP contribution increased nodular dry biomass for the five genotypes in flowering, of which an average rate of 29.58% was observed. Thus, it is at the Sebseb genotype that we noted the lowest rate (19.87%) (Table 4).

Leaf area

The results revealed that the application of P in the form of TCP increased leaf area at all genotypes. This increase was more accentuated when the plants were inoculated by *Bradyrhizobium* strain. Indeed, interaction *Bradyrhizobium*-TCP effect was the most

Genotype	Origin	Taxonomic group	Maturity (days)	100 seed weight (g)	Seed sown ^a weight (g)	
Berrihane	North Algeria	Spanish	90-120	24-29	0.31 ± 0.06	
Boumalek	North Algeria	Spanish	90-120	34-38	0.39 ± 0.02	
Tonga	North Algeria	Spanish	90-120	24-28	0.27 ± 0.03	
Metlili	South Algeria	Virginia	135-140	74-82	0.78 ± 0.04	
Sebseb	South Algeria	Virginia	135-140	79-86	0.86 ± 0.01	

^a The seed weight values before sowing are reported by mean ± standard deviation (SD) of three replicates for each peanut genotype.

Table 2Granulometrical and chemical soil properties.

Sand (g kg ⁻¹)	Silt (g kg ⁻¹)	Clay (g kg ⁻¹)	pH _{H2O}	pH _{KCl}
650 ± 1.2	205 ± 2	145 ± 3	7.75 ± 0.1	6.70 ± 0.2
CE (ds/cm)	$CaCO_3$ (g kg^{-1})	Total-N $(g kg^{-1})$	$P_2O_5 \ (mg\ kg^{-1})$	$K_2O \ (mg \ kg^{-1})$
0.26 ± 0.07	37.67 ± 1.95	1.25 ± 0.16	17.43 ± 1.86	223 ± 7

Data are means and SD of 3 replicates for soil used in this assay.

Table 3 Physicochemical characteristics of water used for irrigation.

pH	CE (µS/cm)	$PO_4^{3-} (mg l^{-1})$	$NO_3^- (mg l^{-1})$	$NO_2^- (mg l^{-1})$	$Ca^{2+} (mg l^{-1})$
8.66 ± 0.09	2576 ± 81.44	0.11 ± 0.03	3.91 ± 0.3	0.05 ± 0.02	441 ± 2.52

Data are means and SD of 3 replicates for water of irrigation.

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