Phosphorus Efficiency of Potato Genotypes

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Dedicated to my parents and my family

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GENERAL ABSTRACT

Potato has high phosphorus (P) fertilizer requirement for optimum growth and yield. However, the low P availability enforces the use of P-efficient genotypes/ cultivars to sustain crop production. The objectives of the present study were to screen potato genotypes for P efficiency, to identify the mechanism and traits associated with P efficiency and to evaluate the contribution of root traits to the predicted P uptake of the genotypes using mechanistic simulation model. To meet these objectives 20 potato genotypes were first screened in two soil experiments at low P (100 mg P kg^{-1} soil) and high P (700 mg P kg^{-1} soil) supply. Based on consistent performance in terms of shoot dry matter yield and relative shoot growth rate, two genotypes (CGN 17903 and CIP 384321.3) were identified as P-efficient and two other genotypes (CGN 22367 and CGN 18233) were identified as P-inefficient. These four genotypes were invesigated both in soil and nutrient solution experiments to identify the mechanism of P efficiency and traits associated with the P efficiency mechanisms. Results of soil experiment showed that P efficiency of genotype CGN 17903 was related to high P utilization efficiency whereas that of genotype CIP 384321.3 was related to both high uptake efficiency in terms of root-shoot ratio and intermediate P utilization efficiency. On the other hand, the P inefficiency of genotype CGN 18233 was related to low P utilization efficiency. With genotype CGN 22367, both low P uptake efficiency (low root-shoot ratio) and intermediate P utilization efficiency contributed to the P inefficiency. Further investigation of mechanism of P utilization efficiency of the genotypes in nutrient solution experiment under three P regimes (10, 45 and 90 µM applied as KH_2PO_4) revealed that the high P utilization efficiency of genotype CGN 17903 under low P supply was related to higher net assimilation rate (NAR). To the contrary, the low P utilization efficiency of genotype CGN 18233 was due to low NAR and this lower NAR was speculated to be due to higher carbon cost of root respiration and/or exudation. With genotype CGN 22367 the intermediate P utilization efficiency could be explained by higher leaf dark respiration rate.

Keywords: net assimilation rate, P efficiency, potato genotypes, root-shoot ratio

KURZFASSUNG

Kartoffeln haben einen hohen Phosphordüngerbedarf, der durch die Verwendung P-effizienter Genotypen verringert werden kann. Die Zielsetzung dieser Arbeit war ein Screening von Kartoffelgenotypen auf P-Effizienz, die Identifizierung von Mechanismen und Eigenschaften, die in Zusammenhang mit der P-Effizienz stehen. Hierfür wurden 20 Kartoffelgenotypen bei niedrigem P (100 mg P kg^{-1} Boden) und hohem P (700 mg P kg^{-1} Boden) Angebot in zwei Topfexperimenten gescreent. Aufgrund von Sprosstrockenmasse und relativer Sprosswachstumsrate wurden zwei Genotypen (CGN 17903 und CIP 384321.3) als P-effizient und zwei Genotypen (CGN 22367 und CGN 18233) als P-ineffizient eingeordnet. In Topfkultur und Nährlösungsexperimenten wurden bei diesen vier Genotypen der Mechanismus der P-Effizienz und die Eigenschaften, die mit dem P-Effizienz Mechanismus verknüpft sind, untersucht. Die Ergebnisse des Versuchs im Boden zeigten für den P-effizienten Genotyp CGN 17903 eine hohe P-Verwertungseffizienz während die P-Effizienz für Genotyp CIP 384321.3 einer hohen Aufnahmeeffizienz und einer mittleren P-Verwertungseffizienz zuzuschreiben war. Die P-Ineffizienz des Genotypen CGN 18233 wurde auf eine niedrige P-Verwertungseffizienz zurückgeführt, die des Genotyps CGN 22367 dagegen auf niedrige P-Aufnahmeeffizienz (geringes Wurzel-Spross Verhältnis) und mittlere P-Verwertungseffizienz. Weitere Untersuchungen zum Mechanismus der P-Verwertungseffizienz wurden in Nährlösungsexperimenten bei drei P Konzentrationen (10, 45 und 90 µM als KH_2PO_4) durchgeführt. Die hohe P-Verwertungseffizienz von Genotyp CGN 17903 bei geringen P- Angebot konnte auf eine hohe Nettoassimilationsrate (NAR) zurückgeführt werden. Im Gegensatz dazu war die niedrige P-Verwertungseffizienz von CGN 18233 einer geringen NAR zuzuschreiben, die vermutlich aus einem höheren Verbrauch von Kohlenstoff durch Wurzelrespiration und/oder Exsudation resultierte. Die mittlere P-Verwertungseffizienz von Genotyp CGN 22367 konnte mit einer höheren Dunkelrespirationsrate der Blätter erklärt werden.

Schlüsselwörter: Nettoassimilationsrate, P-Effizienz, Kartoffelgenotypen, Wurzel-Spross Verhältnis

ABBREVIATIONS

π	Pi
%	percent
$(NH_4)_2SO_4$	ammonium sulphate
℃	degree celsius
µM	micromolar
a.m.	ante meridiem
Al	aluminium
AM fungi	arbuscular mycorrhiza fungi
ATP	adenosine triphosphate
b	buffer power
B	boron
Ca	calcium
$Ca(NO_3)_2$	calcium nitrate
$CaCl_2.2H_2O$	dihydrated calcium chloride
$Ca(H_2PO_4)_2$	calcium phosphate
$CaCO_3$	calcium carbonate
CAL	calcium-acetate-lactate
CGN	centre for genetic resource the Netherlands
CIP	centro internacional de la papa
C_{li}	soil solution P concentration
cm	centimeter
C_{min}	minimum P concentration in soil solution
Co	cobalt
CO_2	carbon dioxide
C_s	P concentration in soil determined by calcium-acetate-lactate method
Cu	copper
$CuSO_4$	copper sulphate
d.m	dry matter
DAT	days after transplanting
D_e	effective diffusion coefficient of P in soil

D_L	diffusion coefficient of P in water
DNA	dioxyribonucleic acid
EDTA	ethylenediaminetraacetic acid
f	impedence factor
F_D	flux by diffusion
Fe	iron
F_M	flux by mass flow
g	gram
g	gravity
H^+	hydrogen ion/proton
H_2CO_3	carbonic acid
H_3BO_3	boric acid
hrs	hours
I_r	maximum uptake rate of root cylinder
I_{rh}	maximum uptake rate of root cylinder plus root hairs
K	potassium
k	root growth rate
K_2SO_4	potassium sulphate
KCl	potassium chloride
kg	kilogram
KH_2PO_4	potassium dihydrogen phosphate
K_m	Michaelis constant
LA	total leaf area
LAR	leaf area ratio
LWR	leaf weight ratio
m	meter
MAFF	Ministry of Agriculture Fisheries and Food
mg	milligram
Mg	magnesium
MgO	magnesium oxide
$MgSO_4.7H_2O$	heptahydrated magnesium sulphate
min	minutes
mL	milliliter

mm	millimeter
mM	millimolar
Mn	manganese
$MnSO_4$	manganese sulphate
Mo	molybdenum
N	nitrogen
$NaMoO_4$	sodium molybdate
NAR	net assimilation rate
NH_4^+	ammonium
NH_4NO_3	ammonium nitrate
$(NH_4)_2SO_4$	ammonium sulphate
nm	nanometer
NMR	nuclear magnetic resonance
P	phosphorus
p.m.	post meridiem
PAR	photosynthetically active radiation
PDW	plant dry weight
pH	negative logarithm of proton concentration
Pi	intracellular phosphate
PROC GLM	procedure for general linear models
PPi	pyrophosphate
PS II	photosystem II
QTL	quantitative trait loci
r_0	root radius
r_{0h}	root hair radius
r_1	mean half distance between neighboring roots
RCBD	randomized complete block design
RFW	root fresh weight
RGR_p	relative plant growth rate
RGR_s	relative shoot growth rate
RHL	mean root hair length per centimeter root
RNA	ribonucleic acid
RNase	ribonuclease

RuBP	ribulose bisphosphate
s	second
SA	total surface area of root cylinder
SAC	surface area of 1 cm root cylinder
SAH	surface area of root hairs found on 1 cm root
SAS	Statistical Analysis System
SDMY	shoot dry matter yield
SDW	shoot dry weight
SLA	specific leaf area
t	time
U	total P content
U_pE	uptake efficiency
U_tE	utilization efficiency
V_0	water flow towards the root
V_{0r}	water uptake rate of root cylinder
W	total amount of water transpired
Zn	zinc
$ZnSO_4.7H_2O$	heptahydrated zinc sulphate
θ	volumetric moisture content
α	alpha

GENERAL INTRODUCTION

1. The potato crop and its P requirement

Potato is the fourth largest food crop in terms of fresh produce after rice, wheat and maize and the world's most widely grown tuber crop (Mandal and Chatterjee, 1993). The cultivated potato (*Solanum tuberosum* L.) requires a high amount of phosphate fertilizer for optimum growth and yield (Alvarez-Sanchez *et al*., 1999; Dechassa *et al*., 2003). The P requirement of potato is two-fold greater than that of tomato and pepper crops (Maynard and Hochmuth, 1997). Fertilizer recommendation for optimum potato yield reaches up to 120 kg ha^{-1} P (MAFF, 2000). However, P requirement of potato depends on the type of variety grown, the processing types demanding more P to allow development of larger tuber size (Moorehead *et al*., 1998).

2. Soil phosphorus status and its availability

Despite its importance for normal plant growth and metabolism, P is one of the least accessible nutrients. Many soils are inherently poor in available phosphorus content (Barber, 1995) although the total amount of P in soil may still be high (Wissuwa and Ae, 1999; Vance *et al*., 2003). This is evident from the extremely low soil solution P concentration (<1 µM) in sandy soils, alkaline soils and highly weathered soils of tropics and sub-tropics (Reisenauer, 1966). Moreover, a large fraction of total soil P is in organic form in many soils and these forms are not directly available to plants (Jungk *et al*., 1993; Richardson, 1994; Smith, 2001; Vance *et al*., 2003). Many of the agricultural soils in the developing countries in particular are P-deficient (Velk and Koch, 1992) and have an unfavourable condition for P availability (Soltan *et al*., 1993). It is estimated that crop productivity is limited by P deficiency on more than 40% of the world arable lands (Vance, 2001). Additionally, world's resources of P are limited (Vance *et al*., 2003).

Unlike nitrate, which readily moves in soil towards the roots via both mass flow and diffusion, phosphate ion is highly immobile in mineral soils. Thus, mass flow

delivers only little phosphate ions (1-5% of plant demand) and the greater portion of required phosphate ions reach the root surface via diffusion (Lambers *et al*., 2006). However, the diffusion coefficient for phosphate ion in soil is very low compared to those for other nutrients (Clarkson, 1981); consequently, plants do not deplete the total volume of the rooted soil layer but only that part of the soil which is in the immediate vicinity of the roots (Föhse and Jungk, 1983).

Phosphorus is commonly bound to iron and aluminium oxides and hydroxides through chemical precipitation or physical adsorption (Kochian *et al*., 2004). As a result of adsorption, precipitation and conversion to organic forms, only 10-30% of the applied phosphate mineral fertilizer can be recovered by the crop grown after the fertilization (Holford, 1997; Gahoonia and Nielsen, 2004b). The rest stays in the soil and may be used by crops in the following years. As a result of low P solubility and desorption, only a small proportion of phosphate ions exist in the soil solution for plant uptake even under optimum P fertilization (Holford, 1997), suggesting that chemical fertilizer application alone is not a cost effective way of increasing crop production in many P-limiting soils (Tilman *et al*., 2002). Therefore, the use of genotypes/cultivars with improved root traits able to unlock and absorb P from bound P resources and/or effectively utilizing the absorbed P is of paramount importance for enhancing the efficiency of P fertilization.

3. Phosphorus efficiency

Phosphorus efficiency is a term that generally describes the ability of crop species/genotypes of a given plant species to give higher yield under P-limiting condition (Graham, 1984). Plant species as well as genotypes within the same species may differ in P efficiency (Föhse *et al*., 1988; Gourley *et al*., 1993; Blair, 1993; Gunes *et al*., 2006; Schenk, 2006). The ability of a crop/genotype to give higher yield under P-limiting condition may be related to: the ability to take up more P from the soil under P-limiting condition (uptake efficiency) or the ability to produce higher dry matter per unit of P in the plant tissue (utilization efficiency) or a combination of both (Gahoonia and Nielsen, 1996).

3.1 P uptake efficiency

For increased P uptake efficiency, plant species/genotypes may use various adaptation mechanisms to gain access to previously unavailable soil P reserves such as through altered root morphology, exudation of chemical compounds into the rhizosphere and association of roots with mycorrhiza (Raghothama, 1999; Vance *et al*., 2003; Lambers *et al*., 2006). Higher P uptake efficiency is usually related to either larger root system size (higher root-shoot ratio) or to higher uptake rate per unit of root length (Föhse *et al*., 1988; Bhadoria *et al*., 2004).

3.1.1 Root morphology

Root architecture

Root architecture refers to the complexity of root system spatial configurations that arise in response to soil conditions (Vance *et al*., 2003) and soil P limitation is a primary effector of root architecture (Williamson *et al*., 2001). Some plant species/genotypes alter the architecture of their root systems under P stress conditions to optimize P acquisition. Adaptations that enhance acquisition of P from the topsoil are important because of the relative immobility of P in the soil, with the highest concentrations usually found in the topsoil and little movement of P into the lower soil profiles (Vance *et al*., 2003). Studies with *Phaseolus* indicated that genotypes that have highly branched root systems and more root apices are efficient in acquiring P. Additionally, P-efficient genotypes also grow lateral roots from the basal roots at an angle that enables them to better explore the upper layers of the soil relatively rich in P (Lynch, 1995; Lynch and Beebe, 1995; Lynch and Brown, 2001). Efficient genotypes develop an architecture that places active roots in regions of the soil more likely to contain available P (Smith, 2001).

Root–shoot ratio

Because of low mobility of phosphorus in the soil, some plant species/ genotypes develop larger root systems that allow a plant to have access to greater soil volume so that higher quantity of soil P can reach the root surface for uptake (Jungk, 2001; Bhadoria *et al*., 2004). Higher root-shoot ratio is often reported for P stressed plants as compared with P sufficient plants (Gaume *et al*., 2001; Bhadoria *et al*., 2002; Bhadoria *et al*., 2004). This is due to severely reduced leaf growth under P stress, which leads to diminished leaf demand for assimilates, consequently causing translocation of photosynthates to the root (Cakmak *et al*., 1994; Ciereszko *et al*., 1996) for better root growth. Preferential root growth thus helps the stressed plants to acquire more P from the ambient environment in response to P stress conditions. Difference in P uptake efficiency between crop species (Föhse *et al*., 1988; Bhadoria *et al*., 2004) and genotypes (Schenk and Barber, 1979; Schenk, 2006) was noticed, which was accounted to difference in root-shoot ratio.

Root hairs

Root surface area alone may not be adequate to feed plants, especially with nutrient of low mobility like phosphorus. Root hairs are tubular outgrowths on the root surface and their formation is mediated by ethylene production (Michael, 2001). Root hairs substantially increase the root surface area for ion uptake (Gahoonia *et al*., 1997; Gahoonia and Nielsen, 1998; Jungk, 2001). Root hairs have a smaller diameter than roots and grow perpendicular to the root axis, which allows better exploration of soil due to enhanced absorptive surface area (Föhse *et al*., 1991; Raghothama, 1999). Root hairs form as much as 77% of the root surface area of field crops (Parker *et al*., 2000). Root hairs are also effective in extending the width of the P depletion zone around the root by increasing the volume of the soil explored for phosphorus (Misra *et al*., 1988; Föhse *et al*., 1991; Smith, 2002).

Some plant species/genotypes are adapted to produce longer and more root hairs under P deficient conditions (Föhse and Jungk, 1983; Bates and Lynch,

1996; Gahoonia *et al*., 1999; Bates and Lynch, 2001; Eticha and Schenk, 2001). However, due to overlapping of nutrient depletion zones developing around the root hairs it is not the density but rather the root hair length which is generally considered to be the most important attribute (Sattelmacher *et al.*, 1994).

Gahoonia and Nielsen (2004a) demonstrated that a barley genotype with a capacity to form longer root hairs (about 1 mm) took up more P, and tended to yield better when P was limiting crop growth compared to genotypes having root hairs half the length (0.5 mm). Gahoonia and Nielsen (1998) reported that root hairs contribute up to 63% of the total P uptake under P deficient condition. Thus, plant species or genotypes of the same species with different root hair length may exhibit different P uptake efficiency (Misra *et al*., 1988; Eticha and Schenk, 2001). However, unlike in soil culture, root hairs play no significant role in P acquisition under hydroponic culture, since diffusion is not a problem (Bates and Lynch, 2000). Root hair growth is genetically controlled and thus traits conferring increased root hair length can be utilized in plant breeding programmes (Föhse *et al*., 1991; Jungk, 2001).

Root radius

Under P deficient condition, plant species/genotypes produce fine roots that allow a contact of larger soil volume per unit of root surface area, thereby increasing P uptake rates (Föhse *et al*., 1991; Gahoonia and Nielsen, 2004b). Thus, plant species/genotypes with thinner roots may be more effective in absorbing soil phosphorus (Gahoonia and Nielsen, 2004b). However, since fine roots tend to turnover more rapidly than coarse roots, the carbon cost of producing finer roots may be higher as these will have to be replaced more frequently (Sattelmacher *et al*., 1994; Gahoonia and Nielsen, 2004b). As a result of this, less consideration is given to select genotypes based on this root morphological trait and to use this trait in breeding programme.

3.1.2 Cluster root formation

Proteoid and dauciform root clusters commonly occurring in plant species belonging to Proteaceae, Retinoaceae, Cyperaceae, Fabaceae and few other families are induced by P deficiency and they are adaptive mechanism to maximize P acquisition from unavailable P resources (Shane and Lambers, 2005; Lambers *et al*., 2006). Both proteoid and dauciform roots are covered with dense mat of root hairs, which markedly increase the surface area of the root system and are also specialized in efficient synthesis and secretion of organic anions (especially citrate and malate) and phosphatases, which help to solubilize insoluble P resources and hydrolyze organic P for plant uptake (Gardner *et al*., 1983; Neumann *et al*., 1999; Playsted *et al*., 2006; Lambers *et al*., 2006). Cluster roots grown under P deficiency exude 20-to 40-times more citrate and malate than those grown under sufficient P (Vance *et al*., 2003).

3.1.3 Association of roots with Arbuscular Mycorrhizae

The vast majority (82%) of higher plant species have the capacity to form a symbiotic association with mycorrhizal fungi (Brundrett, 2002). The degree of dependency on mycorrhizal association under P stress could differ with crop species, those lacking root hairs such as onion being more dependent on the association (Deressa and Schenk, 2008) and also with cultivars (Baon *et al*., 1993; Zhu *et al*., 2001). The symbiotic association of plant roots with arbuscular mycorrhiza (AM) enhances the uptake of nutrients with low mobility like P, especially when the species has a root system that is relatively coarse with few root hairs (Graham and Eissenstat, 1994; Sattelmacher *et al*., 1994). A significant contribution of AM fungi to plant P uptake has been reported especially for soils with low P content and with high P fixing capacity (Marschner and Dell, 1994).

Increased P absorption by mycorrhizal hyphae is related to both increased physical exploration of the soil and modification of the root environment (Bolan, 1991; Smith and Read, 1997; Tinker and Nye, 2000). Enhanced nutrient absorption through physical exploration of the soil due to mycorrhiza is related

to large length of hyphae per unit root length, smaller hyphae radius and larger surface area (Tinker *et al*., 1992). Particularly the ability of hyphae to extend several centimeters out into the surrounding soil allows to expand the effective volume of the soil that the plant can exploit (Smith, 2002).

Arbuscular mycorrhiza fungi also hydrolyze organic phosphate through releasing acid phosphatase into the soil thereby contributing to increased P uptake of the host plant (Tawaraya *et al*., 2005). Arbuscular mycorrhiza hyphae store polyphosphates in their vacuoles, which may be hydrolyzed in the arbuscules and transported as inorganic P into the host plant across the plasma membrane of cells (Smith and Gianninazi-Pearson, 1988). The contribution of AM fungi to P uptake reach up to 77% under low P supply compared to only 49% under high P supply (Thingstrup *et al*., 2000). Furthermore, Deressa and Schenk (2008) reported that fungal hyphae accounted for nearly the whole of predicted P uptake by onion.

3.1.4 Root exudation

Organic acids

Plants growing in an ecosystem low in available P have to obtain P from adsorbed P, sparingly soluble P and organic P complexes. Many plants have developed elegant biochemical mechanisms to solubilize P from insoluble P complexes thereby increasing the pool of P available for uptake (Kirk *et al*., 1999; Neumann and Römheld, 1999; Raghothama and Karthikeyan, 2005). Organic anions such as citrate and malate are the major root exudates released, in response to P deficiency for mobilizing P for plant uptake (Keerthisinghe *et al*., 1998; Neumann and Römheld, 1999; Dechassa and Schenk, 2004).

The range of organic anions released is, however, dependent on the plant species (Ohwaki and Hirata, 1992; Dechassa and Schenk, 2004). Genotypes of the same species may also differ in their ability to exude organic anions and

hence in their ability to mobilize P from sparingly soluble P sources (Dong *et al*., 2004; Corrales *et al*., 2007).

Organic anions mobilize inorganic P through complexing metal cations that bind phosphate and displace phosphate from the soil matrix by ligand exchange (Gerke, 1992; Ryan *et al*., 2001; Jones *et al*., 2003; Raghothama and Karthikeyan, 2005). Besides its role in P solubilization, organic anions exudation also protect the roots of some plants from Al toxicity (Ryan *et al*., 1995; Ma *et al*., 2000) thereby enabling root proliferation and increased foraging capacity for P in acid soils (Smith, 2001).

Acid phosphatase and phytase

A major portion (30-80%) of total P in soil is present in organic forms (Jungk *et al*., 1993; Richardson, 1994; Li *et al*., 1997; Smith, 2001; Richardson, 2001; Vance *et al*., 2003). Half of this is in the form of phytin and its derivatives (Tarafdar and Claassen, 1988; Li *et al*., 1997). This organic-P complex needs to be hydrolysed by enzymatic activities before the inorganic P is released into the rhizosphere for plant uptake. Acid phosphatase and phytase are a group of enzymes produced by plants in response to P stress that can hydrolyse a range of organic-P forms thereby enhancing plant P uptake (Tarafdar and Claassen, 1988; Li *et al*., 1997; Hayes *et al*., 2000; Tarafdar and Claassen, 2001) from unavailable P resources. Acid phosphatases functions both as intracellular (vacuolar) and extracellular (secreted) P salvage systems that catalyze the hydrolysis of P from phosphate-monoesters. The intracellular acid phosphatases play significant role in remobilizing P during senescence and P stress (Plaxton, 2004). Phytases secreted by plant roots into the soil hydrolyze the inositol-phosphate while the intracellular phytase will degrade phytic acid, which is the principal storage form of phosphorus in seeds and pollen for remobilization and use during seedling growth and pollen germination (Li *et al*., 1997).

The amount of phytases and acid phosphatases secreted in response to P deficiency differ between plant species as observed by Li *et al*. (1997).

Moreover, an increased exudation of acid phosphatases was observed in several maize genotypes, except with one genotype under P deficiency, indicating that genotypes may also differ in acid phosphatase activity under P stress (Gaume *et al*., 2001). Similarly, barley genotypes exhibited different extracellular phytase activity under P deficiency (Asmar, 1997).

Moreover, secretion of RNase may degrade the nucleic acids present in decaying organic matter (Plaxton, 2004), which still represent an important source of extracellular P that may be exploited by P stressed plants. Some plant species/genotypes may also release protons into the soil to acidify the rhizosphere condition to enhance P uptake from acid-soluble Ca-phosphate (Neumann and Römheld, 1999; Tang *et al*., 2004).

Others

Induction of high-affinity Pi transporters in roots of P deficient plants also play a crucial role in the acquisition of limited P by some plants (Neumann and Römheld, 1999; Plaxton, 2004). Enhanced expression of high-affinity, plasma membrane-bound Pi transporters in roots and a concomitantly increased P-uptake capacity, was reported as a typical P-starvation response (Dong *et al*., 1999). However, other report indicated that diffusion of P in the soil is the key limiting factor for P uptake and a change in uptake systems have little effect on the plants capacity to acquire P from the soil (Raghothama and Karthikeyan, 2005).

3.2 P utilization efficiency

Besides increased acquisition of soil P, efficient utilization of acquired P is also considered an important adaptation for plant growth on low P soils. Phosphorus utilization efficiency refers to the ability of a plant species/genotype to produce higher dry matter per unit of P absorbed (Blair, 1993).

The mechanism of higher internal P utilization efficiency is not clearly known. However, it may be related to the ability of a plant in releasing inorganic P from

the storage pool (vacuole) to the cytoplasm (cytoplasmic P homeostasis) (Plaxton and Carswell, 1999; Raghothama, 1999) or to selective allocation of P between cytoplasm and vacuole in favour of cytoplasm thereby ensuring sufficient Pi concentration in metabolically active compartments for normal functioning of plant metabolism (Lauer *et al*., 1989a; Lee *et al*., 1990; Raghothama, 1999). Additionally, higher internal P utilization efficiency may also be due to lower metabolic requirement for inorganic P at cellular level under P stress possibly due to the presence of alternative P-independent enzymes/metabolic pathways and/or energy sources (Duff *et al*., 1989; Sattelmacher *et al*., 1994; Plaxton and Carswell, 1999).

3.2.1 Cytoplasmic P homeostasis

The fastest and largest manifestation of P starvation is a decline in intracellular P (Pi) concentration (Natr, 1992). However, plants are still able to maintain cytoplasmic Pi either through effective buffering with vacuolar Pi (Plaxton and Carswell, 1999; Raghothama, 1999) or possibly through selective allocation of Pi between cytoplasm and vacuole to constantly keep sufficient Pi in metablically active compartment (cytoplasm) despite the P stress as confirmed by ^{31}P-nuclear magnetic resonance (NMR) studies (Lauer *et al*., 1989a; Lee *et al*., 1990). During nutritional P limitation, vacuolar Pi is released into the cytoplasm in a regulated manner that correlates with the severity of the Pi stress, ultimately ensuring a relatively constant Pi concentration in the cytoplasm (Mimura *et al*., 1996). The efficiency of this process is, however, dependent on the relative permeability of the tonoplast to Pi (Mimura *et al*., 1990; Schachtman *et al*., 1998) and may vary between different plant species.

Thus, the decline in cytoplasmic Pi, due to absence of effective Pi homeostasis directly affects sugar-phosphate export from the chloroplast (Flügge *et al*., 1980). This situation leads to the decline in Pi levels in the chloroplast stroma and an increase in starch synthesis (Heldt *et al*., 1977; Plaxton and Carswell, 1999). The decline in the stromal Pi can limit the photosynthetic capacity and hence plant growth rate.

There are divergent reports as to how P deficiency results in non-stomatal induced decline of plant photosynthetic capacity. Most common reports state that P deficiency reduces photosynthetic capacity through: (i) directly affceting ATP production (Rao *et al*., 1989), (ii). inactivation of enzymes involved in the ribulose bisphosphate (RuBP) regeneration (Rao and Terry, 1989; Fredeen *et al*., 1990) (iii) inactivating RuBP carboxylase enzyme which catalyzes CO_2 fixation (Lauer *et al*., 1989b), (iv) combined effect of ii and iii (Brooks, 1986; Brooks *et al*., 1988).

High P utilization efficiency (high yield or relative growth rate per unit of P) of a plant species/ genotype under P deficiency can be related to higher net carbon fixation (Fujita *et al*., 2004; Yong-fu *et al*., 2006) achieved possibly through effective cytoplasmic P homeostasis or through selectively allocating more Pi to the cytoplasm. Higher P utilization efficiency under P limiting condition may also be caused due to lower carbon demand for root respiration (Nielsen *et al*., 2001).

3.2.2 The use of P-independent enzymes/pathways in metabolism

In general, little is known about metabolic adaptations of plant respiratory pathways to P deficiency. Alternative use of P-independent enzymes instead of P-dependent ones in glycolysis pathways have been reported as plant respiratory adaptation to P deficiency (Duff *et al*., 1989; Theodorou *et al*., 1992; Theodorou and Plaxton, 1996; Plaxton and Carswell, 1999; Plaxton, 2004). Moreover, the P-independent glycolytic enzymes may also facilitate interacellular Pi recycling, since Pi is the bi-product of the reactions catalyzed by these enzymes, which can be reassimilated into the metabolism of P stressed plants (Plaxton, 2004).

Besides the use of alternative P-independent enzymes, the use of alternative energy source such as the use of pyrophosphate (PPi) in stead of ATP have also been noticed in glycolytic pathways under P stress (Duff *et al*., 1989; Dancer *et al*., 1990; Plaxton and Carswell, 1999). Pyrophosphate is a by-product of anabolic reactions including DNA, RNA, proteins, lipids and

polysaccharide biosynthesis (Plaxton and Carswell, 1999). Plant cytosolic PPi levels are remarkebly insensitive to abiotic stresses such as P-starvation, which elicit significant reductions in cellular ATP pools. The large amount of PPi produced during biosynthesis may be employed by plants to enhance the energy efficiency of several cytosolic processes (Plaxton, 2004).

Active transport of protons into the vacuole by PPi-dependent H^+ pump in the tonoplast can use PPi instead of ATP as an energy donor, which can replace the limited ATP pool under P deficiency (Duff *et al*., 1989; Theodorou *et al*., 1992; Theodorou and Plaxton, 1996; Plaxton and Carswell, 1999).

Plants may also use alternative mitochondrial respiration such as nonphosphorylative pathways that can bypass energy-requiring sites (Theodorou and Plaxton, 1993; Vance *et al*., 2003). This allows continued functioning of the citric acid cycle and respiratory electron transport chain under limited ATP production due to P stress.

3.2.3 Maintenance of cell-division and epidermal cell expansion

One of the most striking effect of P deficiency is a reduction in leaf growth both in terms of leaf number and individual leaf size (Radin and Eidenbock, 1984; Lynch *et al*., 1991; Chiera *et al*., 2002). Leaf initiation at the shoot meristem and its lateral expansion are controlled by the activities of cell division (Chiera *et al*., 2002; Assuero *et al*., 2004) and Pi was reported to play a significant role in cell division activities (Sano *et al*., 1999). However, plants may still differ in tolerance of P stress effect on leave growth mainly due to difference in the ability to maintain cell division (Chiera *et al*., 2002; Assuero *et al*., 2004), leaf epidermal cell expansion (Radin and Eidenbock, 1984) or both (Kavanova *et al*., 2006), under lower tissue P concentration. Alternatively, plants may also retranslocate limited P from older leaves to younger leaves to maintain Pi at levels that permit optimal physiological activities including cell division (Plaxton and Carswell, 1999).

Other possible mechanism of efficient P utilization includes conservation of Pi by replacing membrane phospholipids with non-phosphorus galacto and sulfonyl lipids (Plaxton, 2004).

4. Modeling phosphorus uptake

In general, nutrient uptake is a complex process influenced by many parameters. It depends on desorption, nutrient transport in the soil towards the sorbing root surface, ion transport across root membranes and transport to the shoot through the xylem. The use of models can improve our understanding of these processes (Claassen and Steingrobe, 1999). Mechanistic simulation models calculate nutrient uptake of plants based on the information on the concentration of nutrient in the soil solution, its transport to the roots surface and size, morphology and uptake kinetics of the root system (Claassen and Steingrobe, 1999).

4.1 P transport process in the soil

The model calculates nutrient transport towards the root surface by mass flow and diffusion (Barber, 1962). Mass flow is given as the product of water flow towards the root (V_0), which is equivalent to the amount of water lost through transpiration and the concentration of the nutrient in the soil solution (C_{li}).

$$F_M = V_0 C_{li} \quad \text{.....(1)}$$

Diffusion of nutrient towards the root surface is calculated by Fick's law adjusted to the soil conditions (Nye and Tinker, 1977).

$$F_D = -D_e \frac{\Delta C}{\Delta x} \quad \text{.....(2)}$$

where F_D = flux by diffusion, D_e = the effective diffusion coefficient of P in soil (determination of D_e is described in materials and methods of chapter 2), $\frac{\Delta C}{\Delta x}$ =

the concentration gradient of nutrient available to the plant. The minus sign is a convention which indicates that the flux proceeds down the gradient.

Transport via mass flow is less significant compared to diffusion due to low concentration of P in soil solution (Rengel, 1993). The transport of P to the root surface via diffusion is controlled by the gradient of P concentration that exists between the bulk soil and the soil solution at the root surface. This gradient is generated when the soil solution at the root surface is depleted.

4.2 Kinetics of phosphorus uptake

The uptake kinetics describe the relationship between the influx (I_n) of ion and its concentration at the root surface. For the low concentration range, as found in the soil solution, which is mostly below 1 µmol cm^{-3}, this relationship is a saturation curve that can be described by a modified Michaelis-Menten function (Nielsen, 1972).

$$I_n = \frac{I_{max}(C - C_{min})}{K_m + C - C_{min}} \quad \text{.....(3)}$$

where I_{max} is the maximum influx at high P concentration; K_m is the nutrient concentration at which I_n is half I_{max}, C_{min} is the minimum concentration at which no net influx occurs and C is the concentration in soil solution.

The values of I_{max}, K_m, C_{min} may be different with plant species, genotypes and other factors such as plant P status, plant age and temperature (Nielsen and Schjorring, 1983; Jungk *et al*., 1990; Machado and Furlani, 2004; Li *et al*., 2007).

Phosphorus efficiency can be improved by selecting genotypes with high I_{max}, low K_m and low C_{min} (Gahoonia and Nielsen, 2004b). However, improvement in the efficiency of uptake system is of minor significance for nutrients with low mobility such as P, since transport to the root surface rather than uptake by the root is the most limiting step (Sattelmacher *et al*., 1994; Barber, 1995).

Details of plant and soil parameters used for the model calculation in this study including the way they were determined as well as basic model assumptions are descibed in the materials and methods section of chapter 2 and are thus not addressed in detail in this section.

Reports with several crops demonstrated that genotypes differ in P efficiency. It was also shown that the mechanism as well as traits involved in P efficiency was different among genotypes of crop species as described under some of the plant adaptation mechanisms to P stress discussed in this section. Thus, the over all objectives of this study were

1. to screen potato genotypes for P efficiency

2. to identify the mechanism and traits associated with P efficiency

3. to evaluate the contribution of root traits to the predicted P uptake of the genotypes using mechanistic simulation model

CHAPTER 1

Screening of potato genotypes for phosphorus efficiency

1. Introduction

Potato has a high phosphorus (P) fertilizer requirement for optimum growth and yield (Alvarez-Sanchez *et al*., 1999; Dechassa *et al*., 2003). However, it is estimated that P availability to plant roots is limited in nearly 67% of the cultivated soils causing an important constraint to crop production (Batjes, 1997). To avert this problem, the use of genotypes/cultivars with high P efficiency is an option and makes a better strategy for sustainable potato production in inherently low P soils as well as in soils with high P fixing capacity.

Plant species as well as genotypes/cultivars of the same plant species differ greatly in their ability to tolerate P deficient soils and this difference can be positively exploited. Development of P-efficient cultivars through breeding on the other hand demands the availability of sufficient genotypic variation for P efficiency (Gunes *et al*., 2006). Phosphorus efficiency is defined as the ability of a genotype to produce higher yield compared to other genotypes under P limiting condition (Graham, 1984; Buso and Bliss, 1988; Ozturk *et al*., 2005; Schenk, 2006; Gunes *et al*., 2006). Phosphorus efficiency can arise from the ability of a genotype to acquire P from the soil and accumulate it in the shoots under P limiting condition (uptake efficiency) and/or the ability of a genotype to effectively utilize internal P to produce dry matter (utilization efficiency) (Föhse *et al*., 1988; Blair, 1993; Gourley *et al*., 1993).

According to Gerloff (1977), plant genotypes can be classified into four groups with respect to their response to nutrient deficiency: (1) efficient responders: plants producing high yield at low nutrient level and showing high response to nutrient addition, (2) inefficient responders: plants producing low yield at low nutrient level and showing high response to added nutrient, (3) efficient non-responders: plants producing high yield at low nutrient level but not responding

to nutrient addition, (4) inefficient non-responders: plants producing low yields at low nutrient level and also showing low response to nutrient addition.

To achieve sufficient and useful genetic variation for P efficiency, relatively a large number of genotypes need to be screened. Thus, the aim of this study is to screen 20 wild and cultivated potato genotypes collected from Ethiopia, The Netherlands and Germany for P efficiency so that the P-efficient genotypes can be further closely investigated for mechanisms of P efficiency and identification of relevant traits associated with P efficiency.

2. Materials and methods

2.1 Plant material

Twenty wild and cultivated potato genotypes (Table 1) collected from Ethiopia (Digemegn, Menagesha, Ciro, Zengena, CIP 384321.3, Gorebella and Ambo Local), The Netherlands (CGN 17903, CGN 22367, CGN 18233, CGN 18247, CGN 17829, CGN 18003, CGN 17815 and CGN 18109) and Germany (GLKKS 0081, GLKKS 0557, GLKKS 0872, GLKKS 1263 and GLKKS 1587) were used for the study. Experimental code used to designate each genotype is given in Table 1. In-vitro plantlets of uniform size were obtained by shoot tips culturing in MS medium. To obtain uniform sized in-vitro plantlets of each genotype at transplanting, all the genotypes were propagated on the same day.

2.2 Plant growing in substrate

Eight days old in-vitro plantlets were transplanted into peat substrate for acclimatization, where they were grown under high air humidity. The peat was sieved through 5 mm mesh and 7 g $CaCO_3$ per liter of peat was applied to adjust the pH to 6.7. Each plant received 3 mL solution containing in mg L^{-1}: 50 N, 50 K, 25 P, 20 Mg applied in the form of NH_4NO_3, K_2SO_4, KH_2PO_4, and $MgSO_4.7H_2O$, respectively. Micronutrients were applied in the form of a compound fertilizer (Flory ®10) consisting of 10% MgO, 0.5% B, 0.02% Co, 2% Cu, 3.5% Fe, 0.5% Mn, 0.8% Mo and 0.3% Zn, at the rate of 405 mg L^{-1}.

2.3 Plant growing in soil

A subsoil of Luvisol type derived from loess low in phosphorus content was obtained from a forest area in the suburb of Hannover. The soil was air dried and sieved through 2 mm sieve. $CaCO_3$ (3.2 g kg^{-1} of soil) was added to adjust the pH to 6.3. Similarly, potassium and magnesium were applied at the rate (in mg kg^{-1} soil): 50 K and 40 Mg as K_2SO_4 and $MgSO_4.7H_2O$, respectively. Two P levels were obtained by adding 100 mg P kg^{-1} soil (low P) and 700 mg P kg^{-1} of soil (high P) in the form of $Ca(H_2PO_4)_2$. The soil was filled into plastic pots having a volume of 340 mL and compacted uniformly to the bulk density of 1.38 g cm^{-3}. Ten days before transplanting, the pots were watered to a volumetric water content of 0.23 cm^3 cm^{-3} and kept under room temperature for equilibration.

After 10 days of growth in the peat medium, the seedlings were transplanted to the plastic pots. The pots were watered to a volumetric moisture content of θ = 0.23 cm^3 cm^{-3} throughout the growing period. Two days after transplanting (DAT) to the soil, each plant received N in the form of NH_4NO_3 at the rate of 70 mg $plant^{-1}$. Additional 50 mg N and 30 mg K $plant^{-1}$ were given 10 DAT.

The plants were grown in the controlled growth chamber with a day/night temperature of 20/16°C, relative humidity of 70%/ 80% and light intensity of 200 µmol m^{-2} s^{-1}(PAR), supplied for 16 hours day^{-1}. The plants were harvested 26 and 38 DAT during first and second screening, respectively.

2.4 Harvesting and determination of plant parameters

At harvest, shoots were cut off at the soil surface and shoot dry weight was measured after drying at 65°C for 48 hours. Shoot dry weight at transplanting to the soil was derived from seedling shoot length considering dry matter per cm shoot length of 6 harvested seedlings. The relative shoot growth rate (RGR_s; g g^{-1} day^{-1}) was calculated from the initial and final dry weight assuming linear growth of the crop:

$$RGR_S = \frac{SDW_1 - SDW_0}{(t_1 - t_0) \times SDW_0} \quad \text{.....(1)}$$

where SDW is the shoot dry weight (g) and t is time. Subscripts 0 and 1 refer to time at transplanting and final harvest (days).

2.5 Data analysis

The treatments were arranged in Randomized Complete Block Design (RCBD) with six replications. Data were analysed using the PROC GLM procedure of SAS (SAS Institute INC., Cary, USA). Treatment means were compared according to Tukey test and for all analysis, a significance level α = 0.05 was used.

Table1: Potato genotypes used for the screening experiments

Code of genotypes	Actual Name	Biological status
1	Ciro	Cultivar
2	Digemegn	"
3	CIP 384321.3	"
4	Gorebella	"
5	Menagesha	"
6	Zengena	"
7	Ambo Local	"
8	CGN 22367 *(S. bulbocastanum* subsp*. bulbocastan)*	Wild spp.
9	GLKKS 1587 (Laura)	Cultivar
10	GLKKS 0081	"
11	GLKKS 0557	"
12	GLKKS 0872	"
13	GLKKS 1263	"
14	CGN 17815 *(Solanum demissum)*	Wild spp.
15	CGN 17829 *(Solanum multiinterruptum)*	"
16	CGN 17903 *(Solanum chacoense)*	"
17	CGN 18003 *(Solanum microdontum)*	"
18	CGN 18109 *(Solanum okadae)*	"
19	CGN 18233 (*Solanum tuberosum* subsp.*andigena*)	Landvariety
20	CGN 18247 *(Solanum brevicaule)*	Wild spp.

3. Results and discussion

P efficiency parameters

Phosphorus efficiency is defined as the ability of a genotype to give high yield compared to other genotype under P limiting condition (Graham, 1984; Buso and Bliss, 1988; Ozturk *et al*., 2005; Schenk, 2006; Gunes *et al*., 2006). However, if initial size of seedlings differs between genotypes, relative shoot

growth rate (RGR_s) should preferably be used instead of absolute shoot dry matter yield (SDMY) for ranking genotypes for P efficiency (Caradus, 1983).

3.1 First screening

Plant growth and P efficiency

The high P supply significantly enhanced the SDMY of all genotypes except for genotype 15 (Fig. 1A), where SDMY was higher at low P than at high P supply. Therefore, it was decided not to consider this genotype for the screening purpose. Thus, the RGR_s of this genotype was excluded from Fig.1B. Shoot dry matter yield differed significantly among the genotypes both at low and high P supply. The ranking of genotypes was, however, different at low and high P supply. At low P supply, genotypes 1, 12 and 3 had significantly higher SDMY compared to genotypes 20, 14, 15, 18, 17, 19 and 8. Shoot dry matter yield of the genotypes was on average reduced up to 58% due to P deficiency.

Since genotypes varied in their initial weight consequently affecting the yield, the RGR_s was calculated. Thus, RGR_s given in Fig. 1B followed different pattern from SDMY. Similar to SDMY, the high P supply significantly enhanced the RGR_s of all genotypes (Fig. 1B). The pattern of genotypic ranking was different at low and high P supply. At low P supply, genotypes 12, 11 and 3 had significantly higher RGR_s compared to genotypes 20, 14, 18, 17, 9 and 19. Thus, genotypes 12, 11 and 3 were regarded as P-efficient whereas genotypes 20, 14, 18, 17, 9 and 19 were considered as P-inefficient. On the other hand, at high P supply genotypes 11, 12, 10, 2 and 19 had significantly higher RGR_s compared to genotypes 15, 20, 1, 4, 6, 5, 7, 14, 8, 18 and 17.

According to Gerloff (1977), genotypes with above average SDMY/RGR_s at low P supply are considered as P-efficient whereas genotypes with above average SDMY/RGR_s at high P supply are regarded as responsive. Thus, Fig. 2 demonstrated that genotypes 12, 11, 3, 16, 10 and 13 were both P-efficient and responsive; genotypes 9, 19 and 2 were P-inefficient but responsive whereas genotypes 20, 14, 7, 8, 18 and 17 were P-inefficient and non-responsive.

In the first screening, a total of 9 genotypes (5-efficient) and (4-inefficient) were selected for further evaluation. Genotypes 16, 3, 11, 12 and 1 were selected as the five top P-efficient based on RGR_s at low P supply (Fig. 2). Except for genotype 1, all the genotypes selected as P-efficient were also responsive. On the other hand, genotypes 19 and 2 were selected as responsive but P-inefficient while genotypes 8 and 17 were selected as non-responsive and inefficient. Although they were clearly P-inefficient, genotypes 15 and 20 were not selected, since they were extremely non-responsive to P supply and more importantly due to poor seedling establishment at transplanting leading to only 50% survival of the seedlings. Genotype 14 was also not selected due to severe burning and early dropping of most leaves and unhealthy appearance of remaining leaves at low P supply. Likewise, although it was P-inefficient, genotype 9 was not selected for fearing the high disease symptoms observed with this genotype during experimentation.

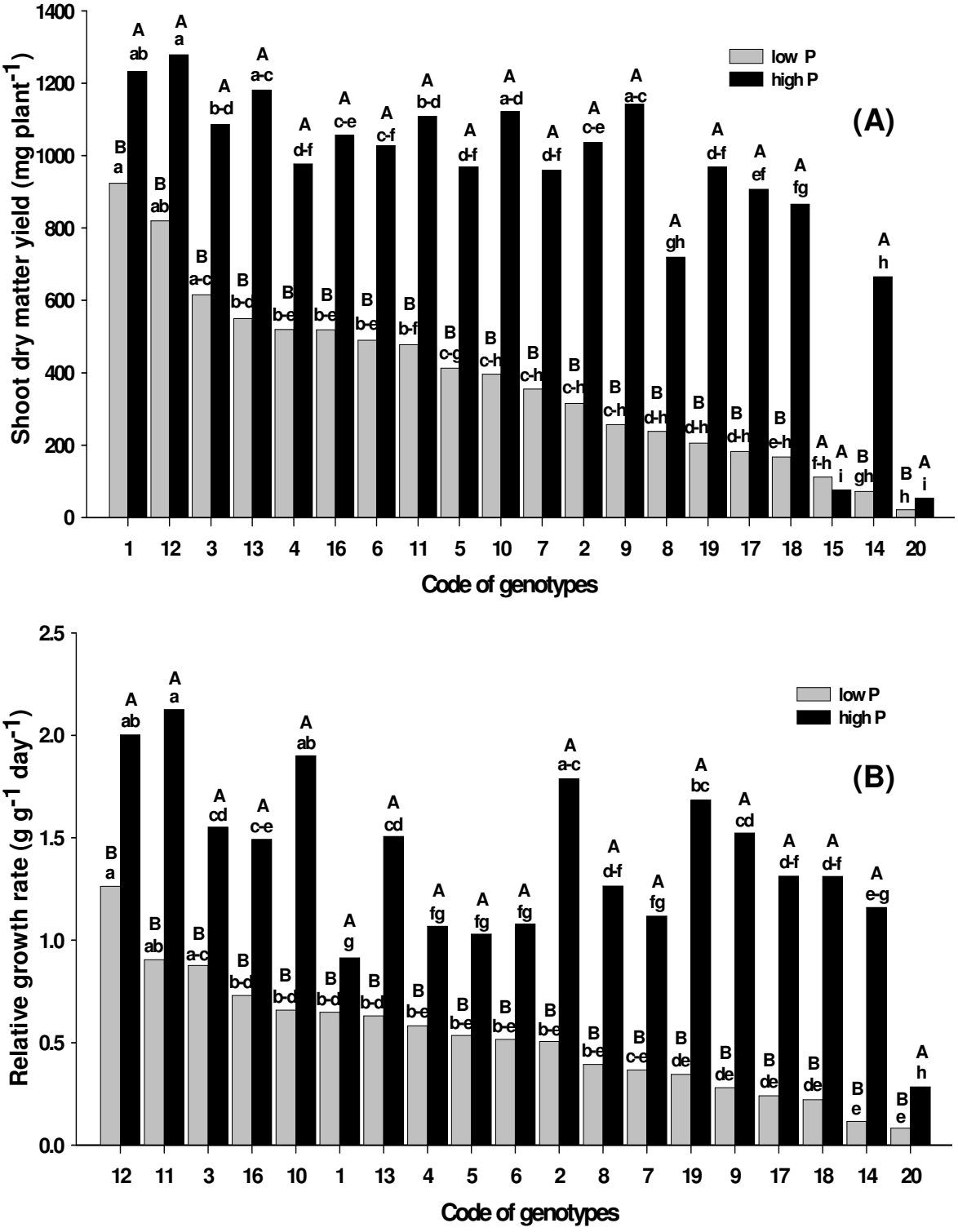

Figure 1: Shoot dry matter yield (A) and relative shoot growth rate (B) of potato genotypes as affected by P supply (different small letters indicate significant difference between genotypes at the same P level whereas different capital letters indicate significant difference between P levels for the same genotype, α= 0.05). See Table 1 for code of genotypes.

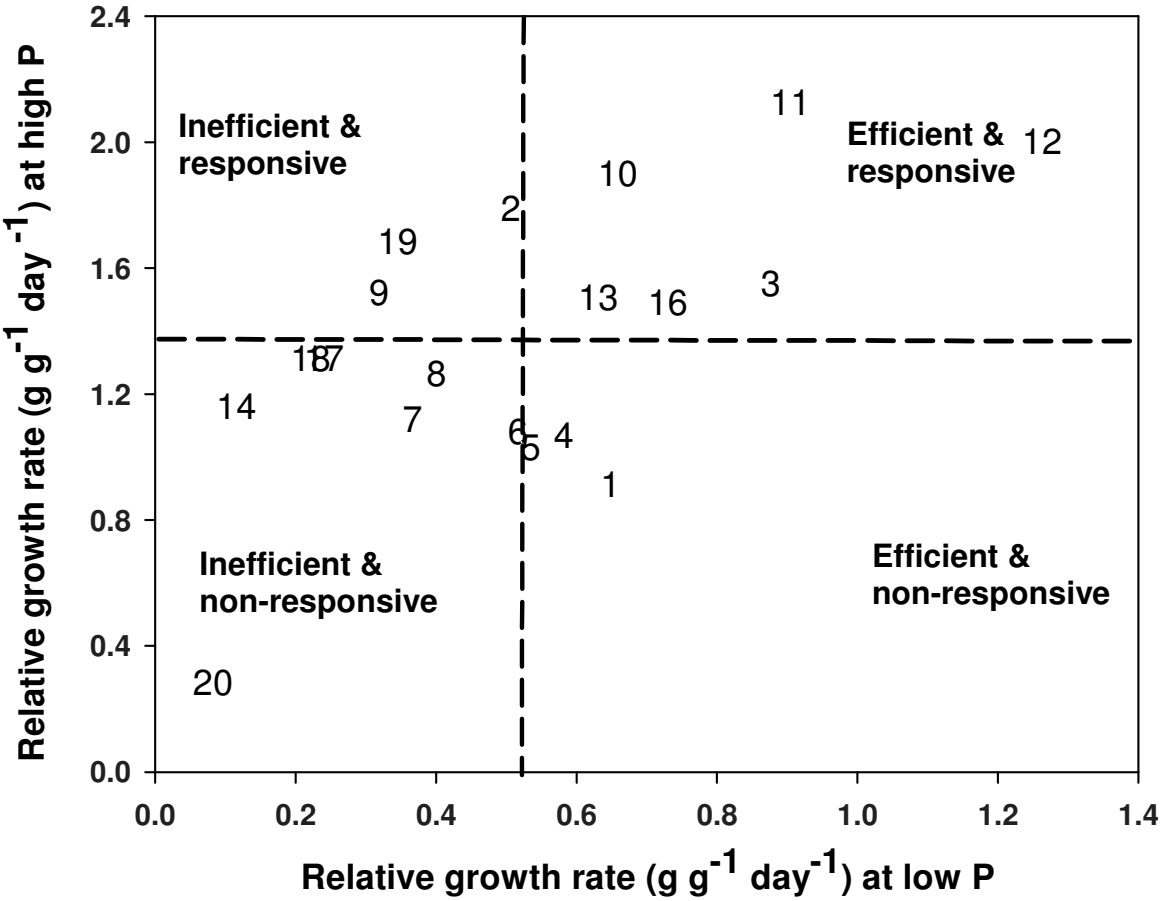

Figure 2: Categorization of potato genotypes for P efficiency and responsiveness based on relative shoot growth rate at first screening (broken lines indicate average relative shoot growth rate). See Table 1 for code of genotypes.

3.2 Second screening

Plant growth and P efficiency

Similar with the first screening, the SDMY at high P supply for all genotypes was significantly enhanced compared to low P supply (Fig. 3A). Phosphorus deficiency on average reduced SDMY by 56% compared to high P supply. The genotypes significantly differed in terms of SDMY both at low and high P supply. Genotypes 16, 12, 3 and 1 had significantly higher SDMY at low P supply compared to genotypes 8 and 19.

As with SDMY, the RGR_s for all genotypes was also significantly increased at high P supply compared to low P supply (Fig. 3B). Relative shoot growth rate of the genotypes was reduced on average by 60% due to P deficiency. Genotypes differed in RGR_s at both low and high P levels. However, the extent of variability was much more pronounced at low P than high P level. Genotype 16, 11 and 3 had significantly higher RGR_s compared to most of the other genotypes at low P level. On the other hand, genotypes 1, 19, 8 and 12 had significantly lower RGR_s compared to genotypes 16, 11 and 3 at low P supply. Except for genotypes 12 and 1, this was consistent with results of the first screening.

Fig. 4 shows that genotypes 16, 11, 17 and 2 were P-efficient and responsive; genotype 3 was P-efficient and non-responsive; genotypes 8 and 19 were P-inefficient and responsive whereas genotype 1 and 12 were P-inefficient and non-responsive. The results of both screening experiments are summarized in Table 2. Comparison of both screening experiments showed that genotypes 16, 11 and 3 were consistently P-efficient whereas genotypes 19 and 8 were consistently P-inefficient based on their RGR_s at low P supply. The same genotypes, however, did not differ much in responsiveness to P supply. Genotypes such as 12, 1, 2 and 17 lack consistency in performance across the experiments (Table 2) and hence could not reliably be selected either as P-efficient or P-inefficient.

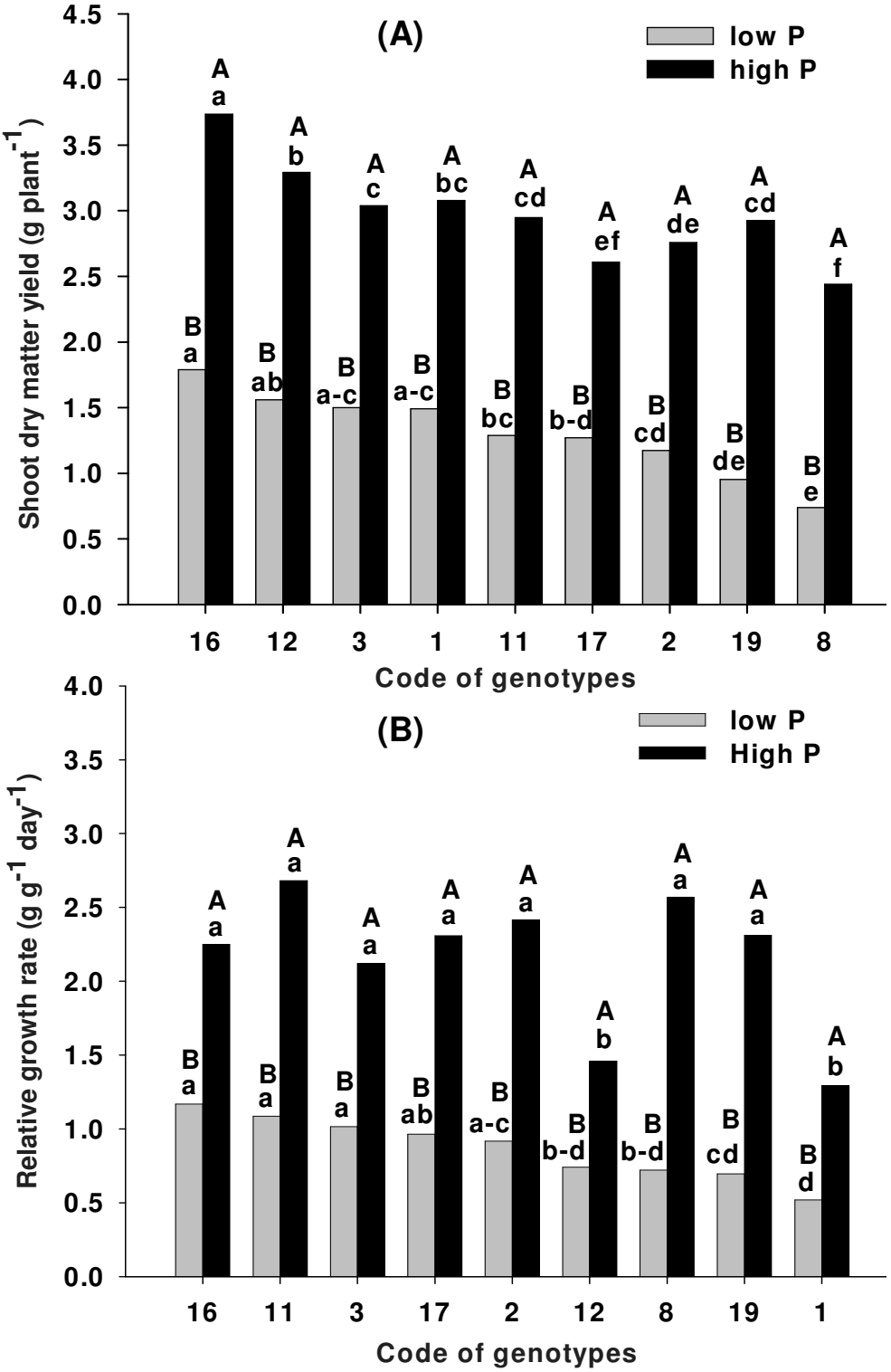

Figure 3: Shoot dry matter yield (A) and relative shoot growth rate (B) of potato genotypes as affected by P supply (different small letters indicate significant difference between genotypes at the same P level whereas different capital letters indicate significant difference between P levels for the same genotype, α= 0.05). See Table 1 for code of genotypes.

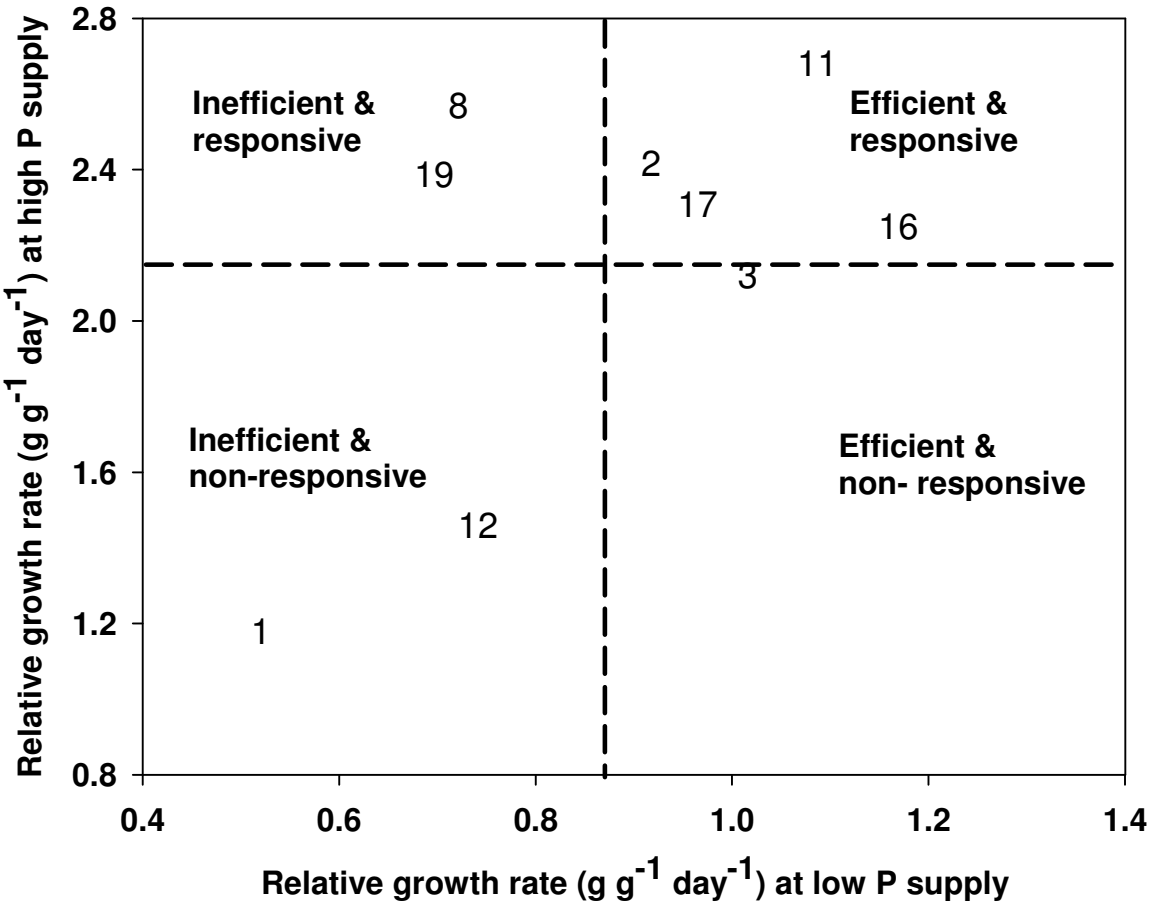

Figure 4: Categorization of potato genotypes for P efficiency and responsiveness based on relative shoot growth rate at second screening (broken lines indicate average relative shoot growth rate). See Table 1 for code of genotypes.

Table 2: Categorization of potato genotypes for P efficiency and responsiveness based on RGR_s at both first and second screening

Code of genotypes	P efficiency and responsiveness category	
	First screening	Second screening
1	Efficient and non-responsive	Inefficient and non-responsive
2	Inefficient and responsive	Efficient and responsive
3	Efficient and responsive	Efficient and non-responsive
8	Inefficient and non-responsive	Inefficient and responsive
11	Efficient and responsive	Efficient and responsive
12	Efficient and responsive	Inefficient and non-responsive
16	Efficient and responsive	Efficient and responsive
17	Inefficient and non-responsive	Efficient and responsive
19	Inefficient and responsive	Inefficient and responsive

4. Conclusions

Since with both screening experiments ranking of genotypes in terms of SDMY and RGR_s followed different patterns due to variability in the initial weight of seedlings, it is suggested that considering the RGR_s is more reliable for evaluating the genotypes for P efficiency and responsiveness than the absolute SDMY. Due to consistent RGR_s results at low P supply, genotypes 16, 11 and 3 are selected as P-efficient whereas genotypes 19 and 8 are selected as P-inefficient.

CHAPTER 2

Genotypic variation of potato for P efficiency and quantification of P uptake with respect to root traits

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Abstract

Potato, an important food crop all over the world, generally requires a high amount of phosphate fertilizer for optimum growth and yield. One option to reduce the need for fertilizer is the use of P-efficient genotypes/cultivars. Thus, twenty wild and cultivated potato genotypes were screened for P efficiency in a growth chamber in soil at low and high P supply. Two efficient and two inefficient genotypes were then selected for investigation of P efficiency mechanisms. The contribution of root traits to P uptake was quantified using a mechanistic simulation model.

For all genotypes, high P supply increased relative growth rate of shoot, shoot P concentration and P uptake rate of roots but decreased root-shoot ratio, root hair length and P utilization efficiency. Genotypes CGN 17903 and CIP 384321.3 were clearly superior to genotypes CGN 22367 and CGN 18233 in terms of shoot dry matter yield and relative shoot growth rate at low P supply, and therefore can be considered as P-efficient. Phosphorus efficiency of genotype CGN 17903 was related to higher P utilization efficiency and that of CIP 384321.3 to both higher P uptake efficiency in terms of root-shoot ratio and intermediate P utilization efficiency. Phosphorus efficient genotypes exhibited longer root hairs compared to P-inefficient genotypes at both P levels. However, this did not significantly affect the uptake rate and the extension of the depletion zone around roots. The P inefficiency of CGN 18233 was related to low P utilization efficiency and that of CGN 22367 to a combination of low P uptake and intermediate P utilization efficiency. Simulation of P uptake revealed that no other P mobilization mechanism was involved, since predicted uptake approximated observed uptake indicating that the processes involved in P transport and morphological root characterstics affecting P uptake are well described.

Keywords: Genotypic variation, P efficiency, P utilization, P uptake, potato genotypes, root-shoot ratio, root hairs

1. Introduction

Potato is an important food crop all over the world. It generally requires a high amount of phosphate fertilizer for optimum growth and yield (Alvarez-Sanchez *et al*., 1999; Dechassa *et al*., 2003). Phosphorus fertilizer requirement of potato is about two-fold higher compared to cereal crops such as wheat and barley and 1/3 higher compared to most vegetable crops (MAFF, 2000). In view of the high P fertilizer input required by the crop, the use of genotypes/cultivars with high P efficiency is an option for sustainable production in low P soils. Phosphorus efficiency is the ability of a genotype to give higher yield under P limiting condition (Graham, 1984). Phosphorus efficiency normally arises either from P uptake efficiency (the ability to take up more P from deficient soil) and/ or P utilization efficiency (the ability to produce more dry matter per unit of P taken up). Phosphorus efficiency mechanisms can differ from genotype to genotype within the same species (Blair, 1993; Gourley *et al*., 1993; Gunes *et al*., 2006). Under P stress conditions, plants may enhance their uptake efficiency through increased root-shoot ratio (Raghothama, 1999; Bhadoria *et al*., 2004; Schenk, 2006), longer root hairs (Bates and Lynch, 2001; Eticha and Schenk, 2001), smaller root radius (Sattelmacher *et al*., 1994; Raghothama, 1999), as well as through release of organic anions and acid phosphatase and association with mycorrhiza (Raghothama, 1999). Physiological root characteristics, describing P uptake according to Michaelis–Menten kinetics such as I_{max}, K_m and C_{min}, are of minor significance in contributing to P uptake efficiency (Barber*,* 1995). Mechanistic simulation models such as that developed by Claassen (1990) can be used to quantify the contribution of morphological root traits to P uptake of plants.

The objectives of this study were to evaluate potato genotypes for P efficiency; to identify the mechanism of P efficiency and to evaluate the contribution of root traits to the predicted P uptake by the genotypes.

2. Materials and methods

2.1 Plant material

Results of four potato genotypes with contrasting P efficiency: CGN 17903 and CIP 384321.3 (P-efficient) and CGN 22367 and CGN 18233 (P-inefficient) are discussed in this paper. These four genotypes were selected out of a total of twenty wild and cultivated potato genotypes collected from Ethiopia (Digemegn, Menagesha, Ciro, Zengena, CIP 384321.3, Gorebella and Ambo Local), The Netherlands (CGN 17903, CGN 22367, CGN 18233, CGN 18247, CGN 17829, CGN 18003, CGN 17815 and CGN 18109) and Germany (GLKKS 0081, GLKKS 0557, GLKKS 0872, GLKKS 1263 and GLKKS 1587/Laura), which were screened for P efficiency based on their relative shoot growth rate and shoot dry matter yield at low P supply in two soil experiments. In both experiments the four genotypes were consistently classified as P-efficient and inefficient.

2.2 Soil preparation

Subsoil of Luvisol type derived from loess low in phosphorus content was air dried and sieved through 2 mm sieve. $CaCO_3$ (3200 mg kg^{-1} of soil) was added to adjust the pH to 6.3. Similarly, potassium and magnesium were applied at the rate (in mg kg^{-1} soil): 50 K, and 40 Mg in the form of K_2SO_4 and $MgSO_4.7H_2O$, respectively. Two P levels were obtained by adding 100 mg P kg^{-1} soil (low P) and 700 mg P kg^{-1} of soil (high P) in the form of $Ca(H_2PO_4)_2$, which resulted in a calcium-acetate-lactate (CAL) soluble P concentration of 66.7 and 479.6 mg P kg^{-1} soil, respectively. The soil was filled into plastic pots having a volume of 340 mL and compacted uniformly to the bulk density of 1.38 g cm^{-3}. Ten days before transplanting, the pots were watered to a volumetric water content of 0.23 cm^3 cm^{-3} and kept at room temperature for equilibration.

2.3 Growing plants and harvesting

Eight days old in-vitro plantlets were transplanted into peat substrate for acclimatization where they were grown under plastic cover. The peat was

sieved and pH was adjusted to 6.7 with $CaCO_3$ (7 g L^{-1}). Each plant received 3 mL solution containing (in mg L^{-1}): 50 N, 50 K, 25 P, 20 Mg applied in the form of NH_4NO_3, K_2SO_4, KH_2PO_4 and $MgSO_4.7H_2O$, respectively. Micronutrients were applied in the form of a compound fertilizer (Flory ®10) consisting of 10% MgO, 0.5% B, 0.02% Co, 2% Cu, 3.5% Fe, 0.5% Mn, 0.8% Mo and 0.3% Zn at a rate of 405 mg L^{-1}.

After 10 days of growth in the peat medium, the seedlings were transplanted to the plastic pots. The pots were watered throughout the growing period to a volumetric moisture content of θ = 0.23 cm^3 cm^{-3} by weighing the pots at each watering considering the increasing weight of the plant. Two days after transplanting (DAT) to the soil, each plant received N at the rate of 70 mg $plant^{-1}$. Additional 50 mg N and 30 mg K $plant^{-1}$ were given 10 DAT.

The plants were grown in a controlled climate chamber with a day/night temperature of 23/16°C, relative humidity of 70%/80% and light intensity of 200 µmol m^{-2} s^{-1}, supplied for 16 hours day^{-1}. The plants were harvested twice, 23 and 38 DAT.

2.4 Determination of plant and soil characteristics

2.4.1 Relative shoot growth rate (RGR_s)

Shoot dry weight at transplanting to soil was derived from seedling shoot length considering dry matter per cm shoot length of 6 harvested seedlings. At harvest shoots were separated from roots and shoot dry weight was determined after oven drying at 65°C for 48 hrs. The relative shoot growth rate (RGR_s; g g^{-1} day^{-1}) was calculated from the initial and final shoot dry weight assuming linear growth of the crop:

$$RGR_S = \frac{SDW_{1,2} - SDW_0}{(t_{1,2} - t_0) \times SDW_0} \quad \text{.....(1)}$$

where SDW is the shoot dry weight (g) and t is time. Subscripts 0,1 and 2 refer to time at transplanting, first harvest and second harvest (days), respectively.

2.4.2 Plant and soil chemical analysis

Shoot P concentration was determined by the vanado-molybdate yellow method according to Gericke and Kurmies (1952). The plant available phosphorus concentration in the soil (C_s) was determined by the CAL method as described by Schüller (1969). For the determination of P concentration in soil solution (C_{li}), about 150 g soil sample (θ = 0.23 cm^3 cm^{-3}) was centrifuged and the P concentration in the supernatant solution was determined with the spectrophotometer by the molybdate blue method according to Murphy and Riley (1962).

2.4.3 Quantifying roots and root hairs

Roots were separated from the soil by washing them under a jet of tap water on a 0.5 mm sieve. After drying between filter papers the total fresh weight was recorded and roots were cut into 1 cm pieces. Three sub-samples, each weighing 0.3 g, were taken for root length determination by using a scanner with WinRHIZO V3.9 software (Regent Instruments, Quebec, Canada).

For the quantification of root hairs, soil was placed under tap water in shallow trays and soaked for few hours. The soil particles settled as sediment and the free floating roots were carefully removed with a pair of pincers. Then roots were cut into pieces of about 1 cm length and kept in plastic vials. Sixty root pieces per replicate were dyed with 1% acid fuchsine solution and then scored into low, medium and high root hair density categories using a microscope (magnification 64x). Root hair length and density for 5 root pieces from each root hair density category was determined by fitting an eyepiece with inscribed square grids each 0.066667 cm size and adjusting the horizontal grids line parallel to the root surface. The number of root hairs crossing the horizontal and vertical grid lines was counted separately for each line. Root hair parameters were computed according to Brewster *et al.* (1976).

2.5 Modeling P uptake

2.5.1 Model description

The mechanistic simulation model described by Claassen and Steingrobe (1999) was used to predict P uptake by the different potato genotypes. The model calculates P uptake based on the assumption that ions are transported to the root surface by mass flow and diffusion using the transport equation of Nye and Marriott (1969) and taken up by the root and root hairs based on Michaelis-Menten kinetics. The model utilizes root morphological characteristics such as root length, root and root hair radius, mean half distance between neighboring roots and root hairs as well as root and root hair surface area to simulate P uptake. A root can only exploit a limited volume of soil ranging from its surface to the half distance towards the neighboring root (r_1). The model assumes that roots are uniformly distributed throughout the soil and that there is equal competition between adjacent roots and root hairs. The nutrient flux at r_1 is assumed to be zero. The soil is assumed as homogenous and isotropic with constant water content (θ) during the calculation period. A constant buffer power (b) was assumed for the calculation (Steingrobe *et al.*, 2000). The root physiological kinetic parameters: maximum uptake rate (I_r /I_{rh}), Michaelis constant (K_m) and minimum concentration (C_{min}) are assumed as constant for model calculation. The data used for the simulation of P uptake are summarized in Table 1.

Table 1: Plant parameters of each potato genotype used for the nutrient uptake model calculation

Physiological root parameters	**CGN 17903**		**CIP 384321.3**		**CGN 18233**		**CGN 22367**	
	Low P	High P	Low P	High P	Low P	High P	Low P	High P
I_r (µmol cm^{-2} s^{-1})	4.84E-7	4.84E-7	3.38E-7	3.38E-7	3.37E-7	3.37E-7	8.40E-7	8.40E-7
I_{rh} (µmol cm^{-2} s^{-1})	2.53E-7	2.53E-7	1.91E-7	1.91E-7	1.99E-7	1.99E-7	4.57E-7	4.57E-7
V_{0r} (cm^3 cm^{-2} s^{-1})	6.68E-7	8.72E-7	2.87E-7	5.52E-7	3.34E-7	6.14E-7	5.73E-7	1.40E-6
Morphological root parameters								
r_0 (cm)	0.011	0.012	0.012	0.013	0.011	0.014	0.009	0.010
r_1 (cm)	0.127	0.133	0.122	0.106	0.124	0.121	0.242	0.145
L_1 (cm)	2559	3080	2823	3023	3127	4133	1288	2414
L_2 (cm)	6318	5868	7096	8959	7014	7144	2357	4709
k (cm day^{-1})	251	186	285	396	259	201	71	153

where I_r = maximum uptake rate of root cylinder; I_{rh} = maximum uptake rate of root cylinder plus root hairs (data for maximum uptake rate (I_r, I_{rh}) were taken from high P treatment); V_{0r} = water uptake rate of root cylinder; r_0 = root radius; r_1 = mean half distance between neighboring roots; L_1 = total root length at first harvest; L_2 = total root length at second harvest; k = root growth rate

2.5.2 Determination of Model parameters

2.5.2.1 Soil parameters

The buffer power (b) was calculated according to Barber (1995):

$$b = \frac{\Delta C_S}{\Delta C_{li}} \quad \text{.....(2)}$$

where C_s = the concentration of P in the soil determined by CAL method and C_{li} = the concentration of P in the soil solution each measured at both first and second harvest and the average value considered for calculating the buffer power.

Thus, the average value of C_s, C_{li} and b was 2.98 µmol cm^{-3}, 0.00182 µmol cm^{-3}, and 1637, respectively for low P and 21.42 µmol cm^{-3}, 0.189 µmol cm^{-3} and 113, respectively for high P.

The impedance factor (f) was calculated according to Barraclough and Tinker (1981) as $f = 1.58\theta - 0.17$ where θ is the volumetric moisture content. Since θ was 0.23 cm^3 cm^{-3} the calculated f value was 0.1934.

Effective diffusion coefficient of P in soil (D_e; cm^2 s^{-1}) was calculated according to Nye (1966) as follows:

$$D_e = \frac{D_L \times \theta \times f}{b} \quad \text{.....(3)}$$

where D_L is the diffusion coefficient of P in water (8.9 x 10^{-6} cm^2 s^{-1}) (Edwards and Huffman, 1959); f is the impedance factor and θ is the volumetric moisture content.

2.5.2.2 Plant parameters

The root growth rate constant (k; cm day^{-1}) was calculated from total root length assuming linear growth as follows:

$$k = \frac{L_2 - L_1}{t_2 - t_1} \quad \text{.....(4)}$$

where L is the total root length (cm $plant^{-1}$) and t is the time of harvests (s). Subscripts 1 and 2 refer to the first and second harvest, respectively.

Mean root radius (r_0; cm): was calculated from root fresh weight (RFW; g) and root length (L; cm) with the formula:

$$r_0 = \sqrt{\frac{RFW}{L \times \pi}} \quad \text{.....(5)}$$

Mean half distance between neighboring roots *(r_1):* was calculated from soil volume (V*;* cm^3) and root length (L*;* cm) as follows:

$$r_1 = \sqrt{\frac{V}{L \times \pi}} \quad \text{.....(6)}$$

Total surface area of root cylinder (SA; cm^2 $plant^{-1}$) was calculated as:

$$SA = 2\pi \times r_0 \times L \quad \text{.....(7)}$$

Surface area of one cm root cylinder (SAC*;* cm^2 cm^{-1}) was calculated as:

$$SAC = 2\pi \times r_0 \times h \quad \text{.....(8)}$$

where h is the length of root cylinder

Surface area of root hairs per cm root length (SAH*;* cm^2 cm^{-1}) was computed as:

$$SAH = RHL \times 2\pi \times r_{0h} \quad \text{.....(9)}$$

where RHL is mean root hair length (cm) per cm root and r_{0h} is root hair radius (cm). For r_{0h} a value of 5 x 10^{-4} was taken from Föhse *et al.* (1991).

P uptake rate *(I_r ;* µmol cm^{-2} s^{-1}) of the root cylinder was calculated as follows:

$$I_r = \frac{U_2 - U_1}{(SA_2 + SA_1)/2} \times \frac{1}{t_2 - t_1} \quad \text{.....(10)}$$

where U is the total P content of the plant shoot (µmol $plant^{-1}$); SA is total root surface area (cm^2 $plant^{-1}$); and t is time of harvests (s). Subscripts 1 and 2 refer to the first and second harvest, respectively.

P uptake rate (I_{rh} ; µmol cm^{-2} s^{-1}) of root cylinder including root hair surface was calculated as:

$$I_{rh} = I_r \times \frac{(SAC)}{(SAC + SAH)} \quad \text{.....(11)}$$

where SAC is the surface area of 1 cm root cylinder (cm^2) and SAH (cm^2) is the surface area of root hairs found on 1 cm root.

Michaelis constant (K_m): A value of 4 µM, which is common for many crops, was taken from Barber (1995).

Minimum P concentration in the soil solution (C_{min}): a value of 0.1 µM, which is common for many crops, was taken from literature (Föhse *et al.,* 1991; Barber, 1995).

Water uptake rate of smooth roots (V_{0r}; $cm^3\ cm^{-2}\ s^{-1}$) was calculated from the water consumption of the plant in analogous procedure with maximum P uptake rate as follows:

$$V_{0r} = \frac{W_2 - W_1}{(SA_2 + SA_1)/2} \times \frac{1}{t_2 - t_1} \quad \text{.....(12)}$$

where W is total amount of water transpired (cm^3); SA is total root surface area ($cm^2\ plant^{-1}$); t is time of harvests (s). Subscripts 1 and 2 refer to the first and second harvest, respectively.

Pots without plants were used to estimate water loss through evaporation. The amount of water transpired was determined by subtracting the amount of water evaporated from the amount of the total water lost from the pots with plants.

2.6 Statistical methods

The treatments were arranged in a Randomized Complete Block Design (RCBD) with 6 replicates per harvest. Data were analysed using the PROC GLM procedure of SAS (SAS Institute INC., Cary, USA). Treatment means were compared according to Tukey test and for all analysis, a significance level $\alpha = 0.05$ was used.

3. Results

3.1 Plant growth

High P supply significantly increased shoot dry matter yield (SDMY) of all genotypes. (Fig.1A). The ranking of genotypes was similar at low and high P supply. However, relative yield difference between genotypes was more pronounced at low P supply: genotype CGN 17903 excelled genotype CGN 22367 by a factor of 2.5 and 1.5 at low and high P, respectively. At low P supply, genotypes CGN 17903 and CIP 384321.3 gave significantly higher SDMY compared to genotypes CGN 22367 and CGN 18233. Similar results were observed during the screening experiment (Chapter 1). Additionally, relative shoot growth rate (RGR_s) was calculated since initial size of seedlings was different between genotypes, affecting the absolute yield. Thus, RGR_s given in Fig. 2 followed another pattern at high P supply: at the first harvest (Fig. 2A) all the genotypes did not differ whereas at the second harvest (Fig. 2B) genotype CGN 22367 was superior to the P-efficient genotypes. However, at low P level genotypes CGN 17903 and CIP 384321.3 were superior to the other genotypes at the second harvest whereas CIP 384321.3 did not differ from CGN 22367 and CGN 18233 at the first harvest. At low P level, ranking of genotypes in terms of SDMY was similar to ranking of the genotypes in terms of RGR_s. Compared to low P supply, RGR_s at high P level at the second harvest increased on average by a factor of 2 for genotypes CGN 17903 and CIP 384321.3 and by a factor of 3.5 for genotypes CGN 22367 and CGN 18233. Similar results were observed during the screening experiment (Chapter 1).

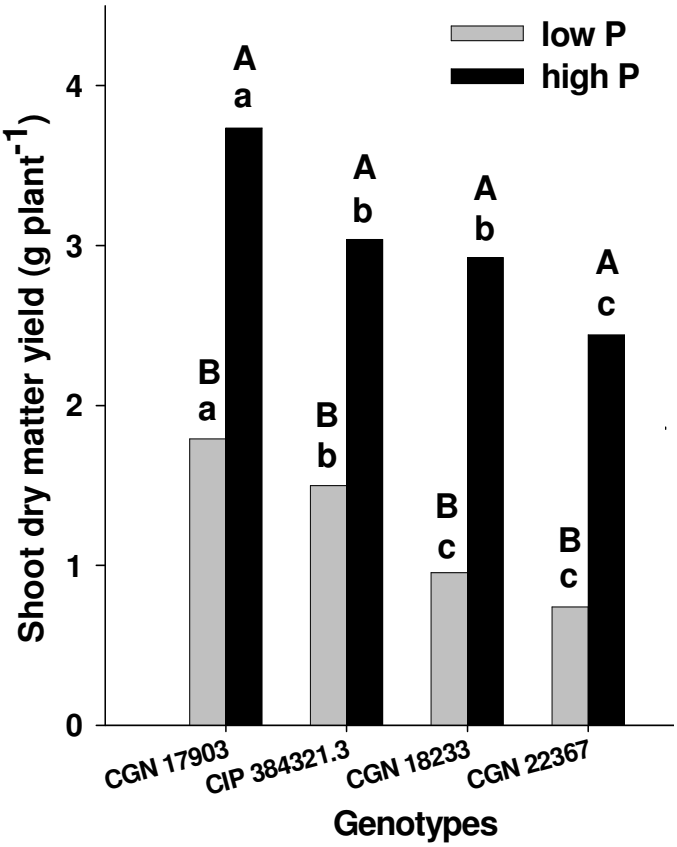

Figure 1: Effect of P supply on shoot dry matter yield of potato genotypes (different small letters indicate significant difference between genotypes at the same P level whereas different capital letters indicate significant difference between P levels for the same genotype, α = 0.05 probability level).

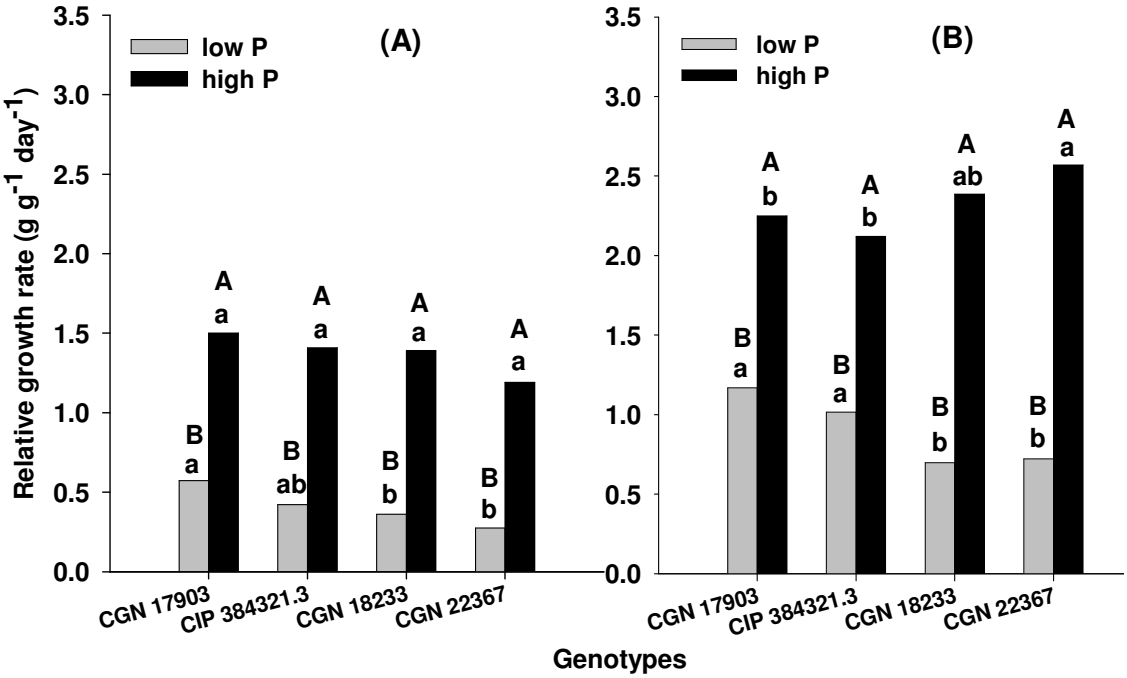

Figure 2: Effect of P supply on relative shoot growth rate of potato genotypes at first harvest (A) and second harvest (B) (different small letters indicate significant difference between genotypes at the same P level whereas different capital letters indicate significant difference between P levels for the same genotype, α = 0.05 probability level).

For three genotypes, root-shoot ratio at low P supply was about two-fold higher than at high P (Fig. 3A) while for genotype CGN 22367 root-shoot ratio did not significantly differ between low and high P. The trend of difference in root-shoot ratio between genotypes was similar for both P levels. Moreover, pattern of root-shoot ratio of the genotypes observed at the first harvest was similar to that of the second harvest except that the absolute value was higher for the first than the second harvest (data for the first harvest not shown). Genotypes CGN 18233 and CIP 384321.3 had higher root-shoot ratio than the other genotypes at both P levels. For all genotypes root hair length increased at low P supply (Fig. 3B). The pattern of ranking the genotypes for root hair length was similar at both P levels. At low P level, genotypes CGN 17903 and CIP 384321.3 had about 0.05 mm longer root hairs compared to the other genotypes.

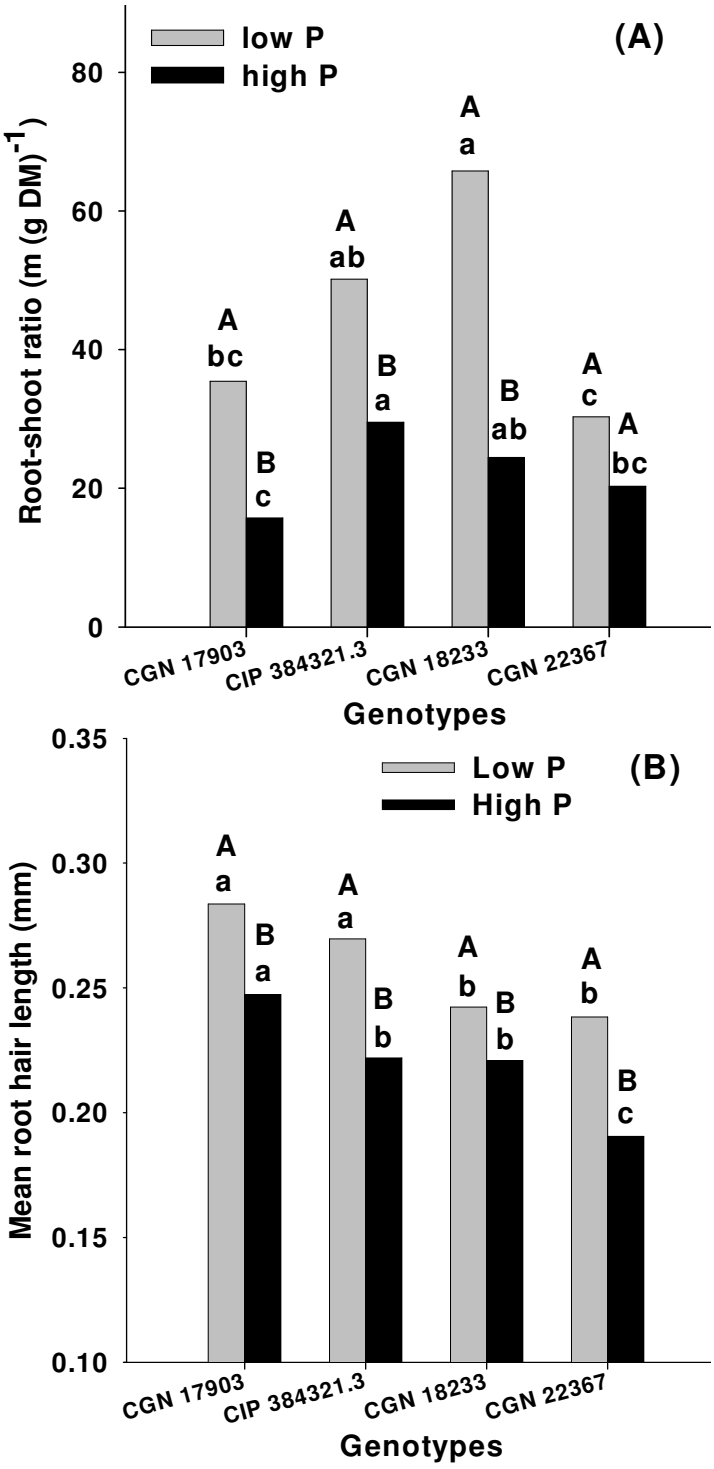

Figure 3: Effect of P supply on root-shoot ratio (A) and root hair length (B) of potato genotypes (different small letters indicate significant difference between genotypes at the same P level whereas different capital letters indicate significant difference between P levels for the same genotype, α = 0.05 probability level).

3.2 Plant P concentration and P uptake rate

Shoot P concentration at low P level was about half of that at high P supply (Fig. 4A). Difference in shoot P concentration among the genotypes did not follow similar trend at low and high P. At high P level, genotype CGN 17903 had lower P concentration compared to the other three genotypes, while at low P level genotype CGN 17903 had lower and genotype CGN 18233 had higher P concentration compared to the other two genotypes. Phosphorus utilization efficiency was higher at low P than at high P supply since it was calculated as a reciprocal of shoot P concentration (Fig. 4B). Consequently, utilization efficiency was higher for genotype CGN 17903 both at low and high P level while genotype CGN 18233 had the lowest P utilization efficiency at low P supply.

At high P supply, P uptake rate was on average 4.5 times higher than at low P level (Fig. 5). Genotypes did not differ in P uptake rate per unit of root length at low P supply. However, at high P supply P uptake rate of genotype CGN 22367 was significantly higher compared to the other genotypes.

3.3 Simulation of P uptake by the mechanistic model

The relationship between model-predicted and experimentally observed P uptake of the potato genotypes under low and high P supply can be seen from Fig. 6A. The P uptake predicted by the model agreed fairly well with the experimentally observed P uptake at both P levels except for genotype CIP 384321.3. Generally, at high P supply a slight over prediction whereas at low P level a slight under prediction was observed. The contribution of root hairs to the predicted P uptake of the genotypes was higher at low P level (Fig. 6B). Root hairs on average contributed about 70% to the predicted P uptake at low P supply and 50% to the predicted P uptake of the genotypes at high P supply. Root hair extended the depletion zone from the root surface by about 0.1 mm over that of the root cylinder at 80% of the initial soil solution P concentration after 15 days of P uptake at low P supply. However, this was not different between genotypes (Fig. 7).

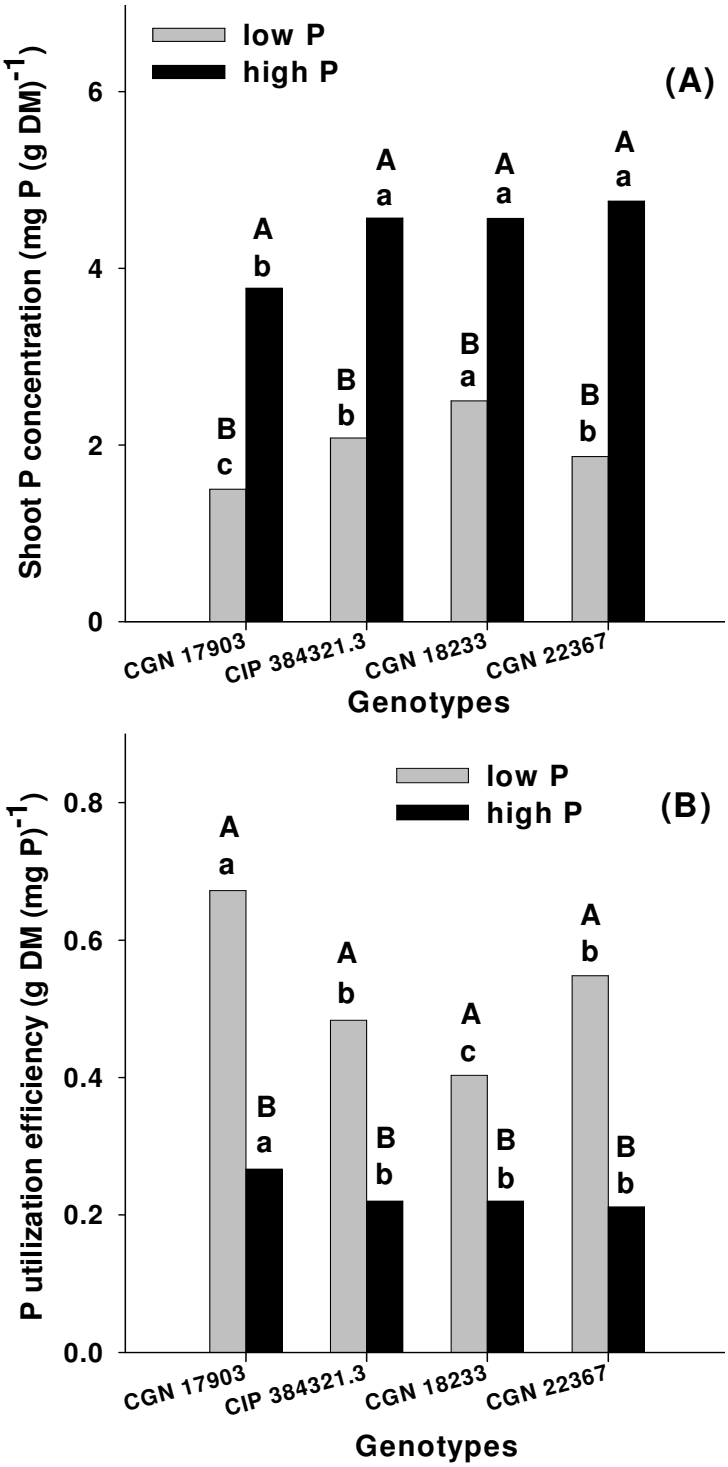

Figure 4: Shoot P concentration (A) and P utilization efficiency (B) of potato genotypes as affected by P supply (different small letters indicate significant difference between genotypes at the same P level whereas different capital letters indicate significant difference between P levels for the same genotype, α = 0.05 probability level).

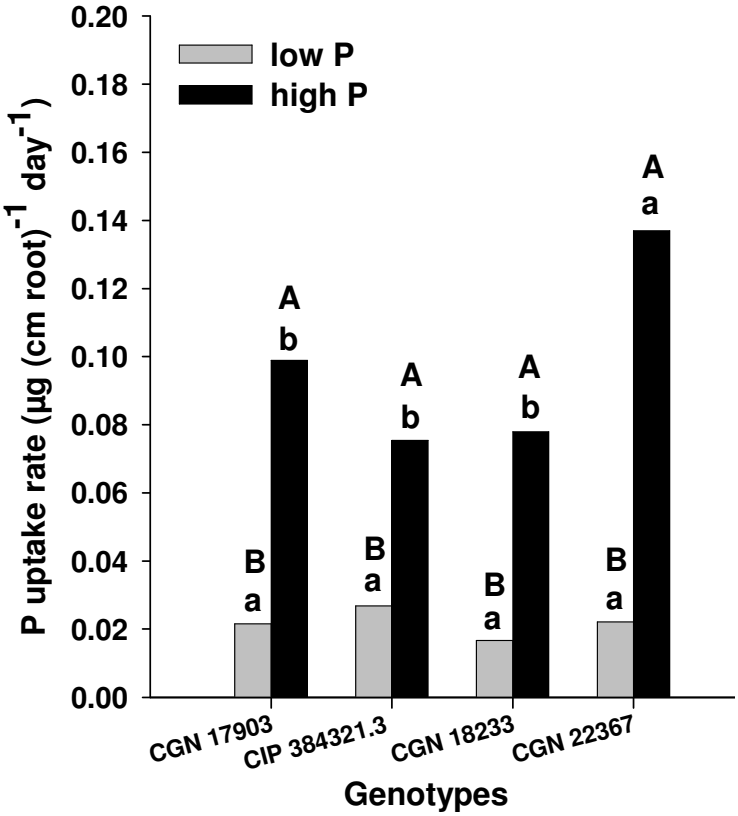

Figure 5: P uptake rate of potato genotypes as affected by P supply (different small letters indicate significant difference between genotypes at the same P level whereas different capital letters indicate significant difference between P levels for the same genotype, α = 0.05 probability level).

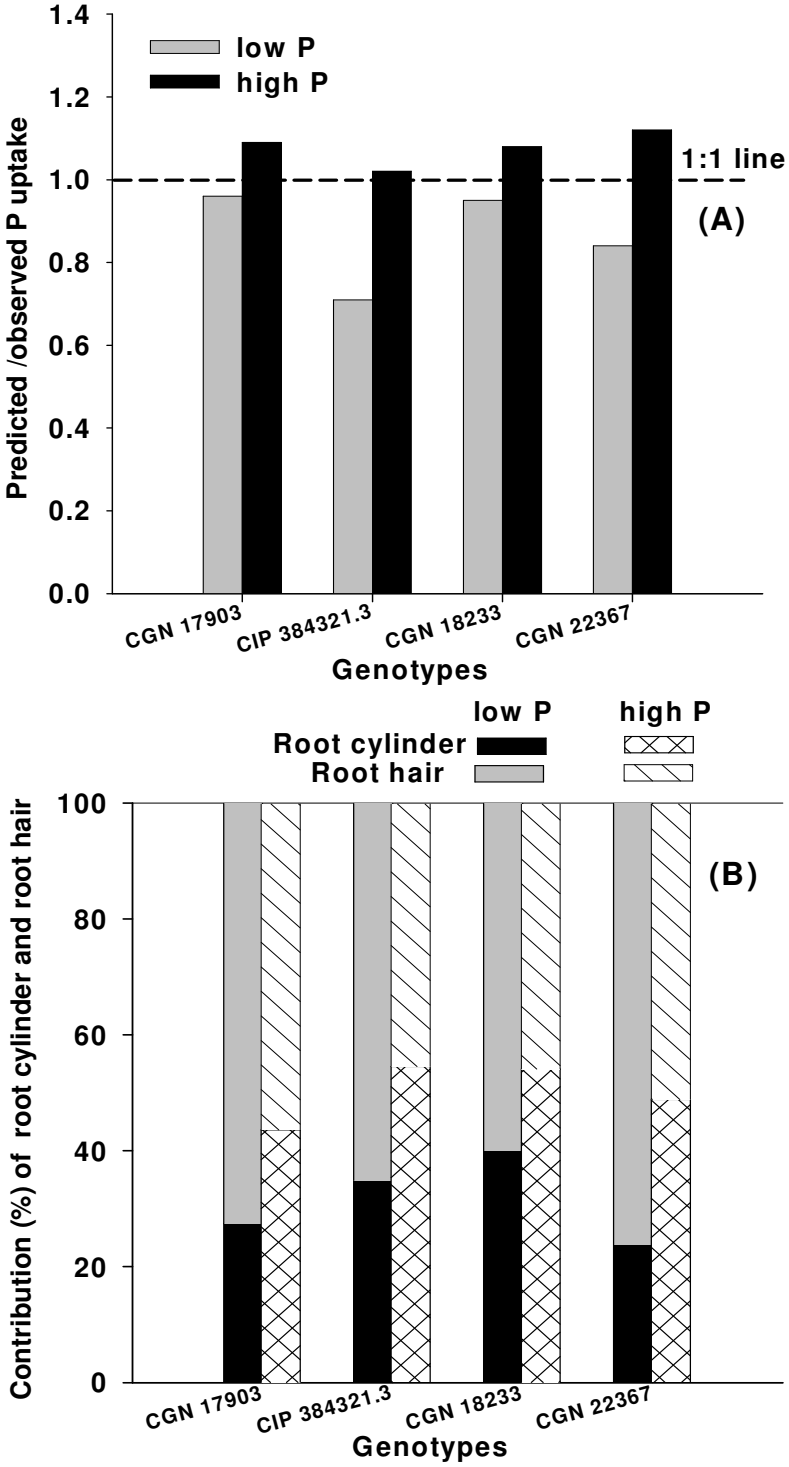

Figure 6: Ratio of predicted and observed P uptake (A) and contribution of smooth root and root hair to predicted P uptake (B) of potato genotypes at low and high P supply.

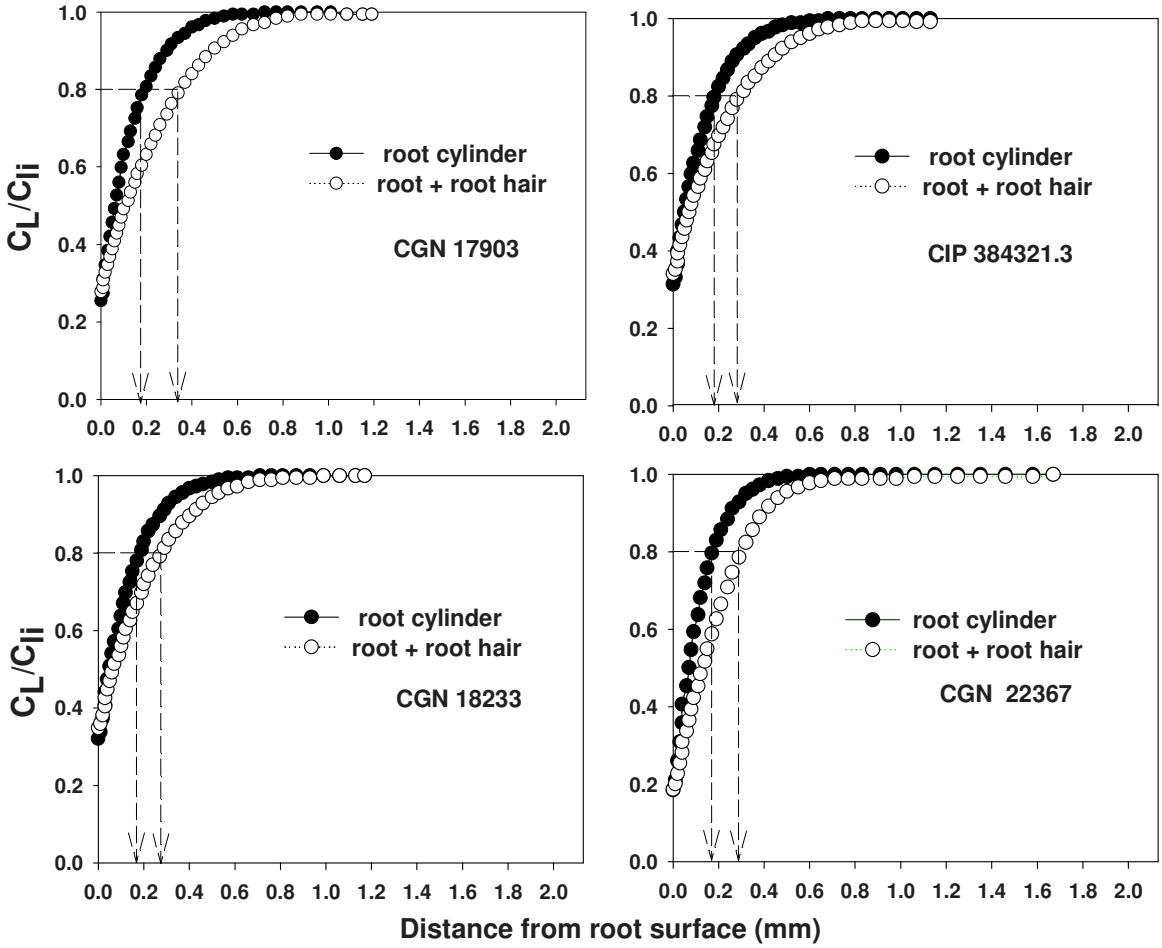

Figure 7: Depletion pattern at the root surface of potato genotypes as simulated for root cylinder and the root cylinder including root hairs after 15 days of P uptake at low P supply (C_L/C_{li} is the *ratio of the actual soil solution P concentration after 15 days of growth to the initial soil solution P concentration).*

4. Discussion

4.1 P efficiency of genotypes

Phosphorus efficiency is defined according to Graham (1984) as the ability of a genotype to produce higher yield compared to other genotype under P limiting condition. Phosphorus efficiency may arise from the ability of a genotype to acquire P from the soil (uptake efficiency) or to utilize P for the production of plant biomass (utilization efficiency) (Blair, 1993; Gourley *et al.*, 1993).

Results of the present study showed that the potato genotypes considerably differed in shoot dry matter yield (SDMY). As expected, low P supply reduced SDMY of the potato genotypes on average by 59% (Fig.1A) and similar observations were reported by Schenk and Barber (1979), Horst *et al*. (1993) and Lynch *et al*. (1991) for corn, wheat and common bean, respectively. Plants grown at low P level showed stunted growth, and had highly reduced leaf size, particularly the P-inefficient genotypes. The reduced leaf size might be related either to reduced epidermal cell production as reported by Assuero *et al*. (2004) for maize and Chiera *et al*. (2002) for soybean or to reduced epidermal cell expansion as reported by Radin and Eidenbock (1984) or to both mechanisms as observed by Kavanova *et al*. (2006) for grass.

Due to variability among genotypes in the initial size of the seedlings at transplanting, relative shoot growth rate (RGR_s) was considered as a more reliable parameter than SDMY to compare genotypes for P-efficiency. However, in terms of both parameters the genotypes had similar ranking at low P supply: the genotypes CGN 17903 and CIP 384321.3 were superior to genotypes CGN 18233 and CGN 22367 (Fig. 1, 2B) while at the first harvest the RGR_s of CIP 384321.3 did not differ from that of CGN 18233 and CGN 22367. This might indicate that efficiency mechanism of CIP 384321.3 developed during plant growth. However, unlike at low P level, ranking of genotypes in terms of SDMY and RGR_s at high P level followed different pattern indicating that the use of RGR_s was more reliable to describe the effect of P supply on growth of genotypes. At the second harvest, the P-inefficient genotypes CGN 18233 and CGN 22367 had higher RGR_s and were more responsive to P supply compared to the P-efficient genotypes (Fig. 2B). At the second harvest, RGR_s of the P-inefficient genotypes was reduced by 72% and that of the P-efficient genotypes by 50% due to low P supply. Classification of the genotypes into four efficiency and response groups according to Gerloff (1977) showed that at the second harvest the P-efficient genotypes CGN 17903 and CIP 384321.3 were non-responders, while the inefficient genotypes CGN 18233 and CGN 22367 were responders to P supply.

4.2 P uptake efficiency

P uptake efficiency is the ability of a genotype to absorb P from the soil under P limiting conditions, which is associated with a higher P uptake rate per unit of root length and/ or with a higher root-shoot ratio (Blair, 1993; Gahoonia and Nielsen, 1996; Bhadoria *et al*., 2004). In this study, the difference in uptake efficiency of the genotypes was not related to difference in uptake rate, since all genotypes had similar uptake rate at low P supply (Fig. 5) although P-efficient genotypes had longer root hairs. This agrees with the observation that root hair contribution to simulated P uptake (Fig. 6B) as well as the extension of the depletion zone from the root surface into the soil (Fig. 7) calculated including root hairs did not notably differ among genotypes.

The second trait related to P uptake efficiency, the root-shoot ratio, was different among genotypes (Fig. 3A). However, the highest root-shoot ratio was observed for the P-inefficient genotype CGN 18233 at both harvests (data for first harvest not shown). Interestingly the P-inefficient genotype CGN 22367 did not respond to low P supply by increasing root-shoot ratio at both harvests. Of the two P-efficient genotypes, CIP 384321.3 had an increased root-shoot ratio comparable with that of CGN 18233 and hence a high P uptake efficiency whereas CGN 17903 showed a lower root-shoot ratio. These diverse results suggest that besides root-shoot ratio, as indicator of P uptake efficiency, P utilization efficiency might also be relevant for explaining P efficiency. In contrast, for maize and soybean P efficiency of genotypes was related to P uptake efficiency in terms of high root-shoot ratio (Schenk and Barber, 1979; Pan *et al*., 2008).

At low P, the root-shoot ratio increased by factor of two compared to high P supply. This observation is in agreement with results reported in literature (Gaume *et al*., 2001; Bhadoria *et al*., 2002; Bhadoria *et al*., 2004). The higher root-shoot ratio observed at low P supply was due to the severely reduced shoot growth (Fig. 1A) compared to root growth (data not shown) as also observed by Bhadoria *et al*. (2002). It is discussed that the severely reduced leaf growth at low P supply leads to diminished leaf demand for assimilates resulting in enhanced translocation of photosynthates to the root (Cakmak *et al*.,

1994; Ciereszko *et al*., 1996) for better root growth. Preferential root growth thus helps the stressed plants to acquire more P from the ambient environment in response to P stress conditions.

4.3 Quantification of P uptake with respect to root traits

The fairly good agreement between experimentally measured and model predicted P uptake (Fig. 6A) at both P levels indicated that the major processes involved in P transport and root characteristics of the genotypes affecting P uptake were well described. These characteristics include root morphological traits such as root growth rate, root radius, mean half distance between neighboring roots and physiological root characteristics such as (I_r/I_{rh}), K_m and $C_{min.}$ However, the under prediction (about 30%) for genotype CIP 384321.3 was observed at low P level. This could not be due to additional P mobilization mechanism, which is not considered by the model, since the P uptake rate per unit of root length of this genotype was in a similar range with that of the other genotypes. The possible reason accountable for the under prediction of P uptake by this genotype could be that some of the physiological root kinetic parameters taken as common for most crops such as K_m or C_{min} (4 µM and 0.1 µM, respectively) might not be representative for this cultivar. Sensitivity analysis for these two physiological root parameters revealed that lowering the K_m value by half resulted in a predicted P uptake similar to the observed value. To the contrary, increasing or decreasing C_{min} by a similar factor did not affect the predicted P uptake. Therefore, the K_m value of this genotype might have been lower than what was assumed for the model calculation. Low K_m values were reported to be indicative of better P uptake efficiency (Machado and Furlani, 2004). Reports by Nielsen and Schjorring (1983), Machado and Furlani (2004) and Li *et al*. (2007) revealed that these parameters differed among barley, maize and rice cultivars, respectively.

According to the simulation results, the contribution of root hairs to the total predicted P uptake was on average 70% at low P and 50% at high P. Not far from our observation, Dechassa *et al*. (2003) reported a root hair contribution of

about 60% in potato at a soil P level of 124 mg kg^{-1} soil, which is more or less similar to our low P level (100 mg P kg^{-1} soil).

4.4 P utilization efficiency

P utilization efficiency refers to the ability of a genotype to produce higher dry matter per unit of P absorbed (Blair*,* 1993) and it is calculated as reciprocal of shoot P concentration. In the present investigation, P utilization efficiency at low P supply was higher than that at high P supply. This is expected and was observed previously in other investigations (Akhtar *et al*., 2008a, b). At low P supply, the P utilization efficiency was the highest for P-efficient genotype CGN 17903, and intermediate for genotypes CIP 384321.3 and CGN 22367 whereas it was the lowest for P-inefficient genotype CGN 18233 (Fig. 3B). Thus, P inefficiency of genotype CGN 18233 is related to low P utilization efficiency, since this genotype was not inferior in P uptake efficiency but even superior in terms of root-shoot ratio. On the other hand, P efficiency of genotype CGN 17903 can be explained primarily by high P utilization efficiency, since it was not superior in P uptake efficiency but even inferior with regard to root-shoot ratio. For *Brassica* cultivars, Akhtar *et al*. (2006, 2008a, b) observed that P-efficient cultivars had the highest P utilization efficiency compared to P-inefficient cultivars. Clark (1983) also reported that P-efficient sorghum genotypes had higher P utilization efficiency than P-inefficient genotypes.

The mechanism of higher internal P utilization efficiency is not clearly known. However, it could be related to a better ability in releasing inorganic P from the vacuole to the cytoplasm (cytoplasmic P homeostasis) or possibly through selective allocation of P between cytoplasm and vacuole in favour of cytoplasm thereby ensuring a relatively constant Pi concentration in metabolically active compartments for normal functioning of plant metabolism (Lauer *et al*., 1989a; Lee *et al*., 1990; Plaxton and Carswell, 1999; *Raghothama*, 1999) or to lower metabolic requirement for inorganic P at cellular level for maintaining normal metabolic activities (Sattelmacher *et al*., 1994). These could be (i) alternative use of P independent enzymes and/or energy sources instead of P dependent ones in metabolic pathways (Duff *et al*., 1989; Plaxton and Carswell, 1999) (ii)

ability to maintain cell division at shoot meristems at lower tissue P concentration leading to maintenance of optimum leaf number per plant (Lynch *et al*., 1991; Chiera *et al*., 2002). (iii) ability to maintain leaf epidermal cell expansion at lower P concentration leading to relatively larger individual leaf area (Radin and Eidenbock, 1984; Kavanova *et al*., 2006). As a consequence of larger leaf area, there would be an interception of more light leading to a better shoot biomass accumulation under low P supply (Plenet *et al*., 2000). High P utilization efficiency might also be related to the ability to maintain a higher photosynthetic rate under P deficiency as observed by Yong-fu *et al*. (2006) for rice genotypes.

5. Conclusions

Potato genotypes investigated in the present study differed significantly in RGR_s and SDMY at low P supply and this difference was related to both P uptake and P utilization efficiency. Superiority of genotype CGN 17903 is related to high P utilization efficiency whereas that of genotype CIP 384321.3 was related to both uptake efficiency in terms of higher root-shoot ratio and intermediate P utilization efficiency. Phosphorus uptake efficiency alone may not necessarily lead to P efficiency as observed for genotype CGN 18233, which had the highest root-shoot ratio but the lowest P utilization efficiency. Therefore, selection of genotypes with both efficiency mechanisms appears to be the most promising approach for a successful breeding program.

CHAPTER 3

Genotypic difference of potato in carbon budgeting as a mechanism of phosphorus utilization efficiency

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GENERAL DISCUSSION

1. Soil phosphorus management and sustainable crop production

Sustainable crop production aims at maintaining high crop yield without adversely affecting ecosystems to meet the need of current as well as future generations (Tilman *et al*., 2002). Since phosphorus in agriculture is the second most growth limiting macronutrient after nitrogen, its proper management in soil contributes significantly to sustainable crop production. In such soils where yield is limited because of inherent low P concentration (P deficient soils), application of relatively higher amount of mineral P fertilizers is the only way to enhance soil available P status to a target value in a long run that can sustain high crop yield. However, once the target value is reached, the available soil phosphorus concentration can be kept at a level that can sustain high crop yield through maintenance fertilization (replacing only the P removed from the field along with the harvested crops).

The P contained in crop residues left in the field can be recycled by incorporating the residues into the soil whereas part of P in crop residues feed to livestock can be returned back to the soil in the form of manure and also as bone meal. The mineralization of such organic P sources can occur through the action of microorganisms and plants exuding phosphatases and phytases. However, the P removed along with cereal grains, other edible vegetable parts and livestock products such as milk and meat used for human consumption need to be replaced through mineral P fertilizer application. Therefore, under condition where P removed from the soil by harvested crop can be returned as crop residues and manures, the amount of mineral P fertilizer required for maintenance fertilization becomes less. In a nutshell, regular application of mineral P fertilizers for crop production and incorporation of crop residues after harvest instead of removing from field can reduce nutrient mining and contribute to sustainable crop production.

2. Problems of mineral P fertilization

The major portion (80-90%) of mineral P fertilizers applied to the soil can not be absorbed by plants due to adsorption to Fe oxides/hydroxides, Al hydroxides as well as to Ca and Mg carbonate surfaces and due to chemical precipitation resulting in the formation of sparingly soluble Fe-phosphates (strengite) and Al-phosphates (variscite) in acid and Ca-phosphates in alkaline soils. Moreover, the applied mineral P fertilizer may also possibly be transformed to organic form, a process known as immobilization (Holford, 1997; Mengel and Kirkby, 2001). Thus, the mineral P fertilizer recovery of crops during the year of application is usually very low (less than 20%).

Mineral P fertilizer recovery of crops can be improved through proper method of P fertilizers application. In soils that have a high P fixing capacity in unavailable forms, band application (where P is concentrated in a narrow zone) enhances P fertilizer recovery compared to broadcasting, since concentrating P in a small soil volume saturates the P binding sites and lowers the buffering capacity of the soil thereby increasing mobility of phosphate ion into the soil solution and its further diffusion toward the root. Band application especially of mono-and di-ammonium phosphate fertilizers enhance root proliferation due to both N and P effect, consequently improving the P uptake capacity of plants. Similarly, in alkaline soils having a high capacity to precipitate P as calcium phosphate, addition of organic materials such as farmyard manure along with mineral P fertilizers improve P solubilization through microbial activities such as excretion of organic anions, and H_2CO_3 formed due to respiration as well as H^+ released by plant root induced by NH_4^+ uptake, all of which result in acidification of the rhizosphere. Besides rhizosphere acidification, the organic anions as well as the H_2CO_3 may also play role in desorption of phosphate ions adsorbed to Ca and Mg carbonates, through ligand exchange reactions. This enhances P availability contributing to improved mineral P fertilizer recovery by crops.

3. P-efficient cultivars and their role in sustainable crop production

Phosphorus efficiency, which is the ability of a genotype/cultivar to produce high yield under P limiting condition (Graham, 1984), can be attained through

improved P uptake efficiency (the ability to take more P from the soil under P limiting condition) and/or through improved P utilization efficiency (the ability to produce higher dry matter yield per unit of P taken up) (Gahoonia and Nielsen, 1996). Thus, P-efficient cultivars produce reasonably high yield in low P soils through either ways and thus can reduce mineral P fertilizer input requirement in agricultural production.

Phosphorus uptake efficient cultivars may contribute to sustainable crop production by producing reasonably high yield under P deficient condition due to their ability to exploit greater soil volume for accessing more P through producing larger root system (higher root-shoot ratio), longer root hairs or via forming association with mycorrhiza. Such cultivars may also enhance the applied mineral P fertilizers recovery and improve P availability, since they may be adapted to mobilize mineral P fertilizers fixed by the soil after application through exuding organic anions and protons. Additionally, P uptake efficient cultivars may also be able to mineralize organic P sources (including those of plant and microbial origin) by releasing acid phosphatases, phytases and/or RNase thereby increasing soil available P to sustain high yield. Thus, P uptake efficient cultivars are able to produce high yield at relatively low soil P status which can be reached by applying less amount of mineral P fertilizer.

On the other hand, P utilization efficient cultivars produce high yield per unit of absorbed P under P deficient condition, since they have low internal P demand for normal metabolic activities and growth and hence have low requirement for mineral P fertilizer inputs to produce reasonably high yield. Moreover, they remove less P from soil during growth and therefore the quantity of P removed along with the harvestable parts of the crop would obviously be less, consequently reducing the quantity of mineral P fertilizer inputs required for maintenance fertilization.

4. Genetic diversity of potato genotypes for P efficiency

The results of the screening experiments (Chapter 1) showed that there is considerable genetic diversity of potato for P efficiency. Phosphorus efficient as well as inefficient genotypes were observed both among the wild and cultivated

types as evaluated in terms of both high shoot dry matter yield and high relative shoot growth rate under P limiting condition (Chapter 1). Overall speaking, the cultivated types seem more P-efficient than the wild types. Besides for P efficiency, the genotypes were also genetically diverse in terms of responsiveness to P supply. The presence of significant genotype*P level interaction ($P<0.001$) indicated that ranking of genotypes for P efficiency and responsiveness was different.

Among the cultivated types, genotype CIP 384321.3 whereas among the wild types, genotype CGN 17903 were consistently P-efficient as evaluated in terms of both high shoot dry matter yield and high relative shoot growth rate at low P supply in both screening experiments. The consistently P-inefficient genotypes were the wild type CGN 22367 and a land variety CGN 18233. The inconsistency in case of some genotypes have been related to shortcomings of the classification method described by Gerloff (1977), since the mean values used to classify the genotypes to either of the P efficiency or P response category were changing depending on the number of genotypes classified and composition of P-efficient and P-inefficient genotypes in each screening experiments.

5. Mechanisms and traits contributing to P efficiency of selected potato genotypes

As described earlier, P efficiency can be achieved due to uptake efficiency and/ or utilization efficiency. In the current study, the P efficiency for the genotype CGN 17903 was related exclusively to higher P utilization efficiency (Chapter 2 and 3), whereas for the genotype CIP 384321.3, it was related to both higher P uptake efficiency in terms of root-shoot ratio and intermediate P utilization efficiency (Chapter 2).

Genotype CGN 17903 showed the highest P utilization efficiency compared to the other selected genotypes (Chapter 2 and 3), which led to the highest relative growth rates (RGR_s/RGR_p) of this genotype under low P level. Similarly, Akhtar *et al*. (2006, 2008a,b) also reported P efficiency of some *Brassica* cultivars to have been due to higher P utilization efficiency. Results of the

present study revealed that genotype CGN 17903 had higher net assimilation rate (NAR), which was the cause for high P utilization efficiency of this genotype. However, the higher NAR of this genotype was not related to higher net photosynthetic rate since this did not differ between the genotypes. Unlike the present observation, Fujita *et al*. (2004) and Yong-fu *et al*. (2006) accounted P efficiency of pigeon pea and rice genotypes, respectively to higher net photosynthetic rate of the P-efficient genotypes compared to the inefficient ones.

On the other hand, the P-efficient genotype CIP 384321.3 showed both higher P uptake and intermediate P utilization efficiency. The P uptake efficiency of this genotype was related to higher root-shoot ratio (Chapter 2). Similarly, higher root-shoot ratio has been accounted for enhanced P uptake efficiency with maize, cowpea and soybean genotypes (Schenk and Barber, 1979; Krasilnikoff *et al*., 2003; Pan *et al*., 2008). However, this genotype did not mobilize P since uptake rate per unit root length was in a similar range with that of other genotypes (Chapter 2). To the contrary, studies with other crop species revealed that P uptake efficiency of barley, cowpea, common bean, maize, rape, rice, soybean and wheat genotypes was also related to their P mobilizing capacity (Table 1).

The P inefficiency of genotype CGN 22367 was due to both lower P uptake efficiency (related to low root-shoot ratio) and intermediate P utilization efficiency (Chapter 2 and 3). The results from an investigation of traits related to P utilization efficiency in a nutrient solution experiment revealed that intermediate P utilization efficiency of this genotype was not related to lower net photosynthetic rate but to higher leaf dark respiration rate (Chapter 3). So far there is no literature report which supports the present observation. On the other hand, although genotype CGN 18233 had a higher P uptake efficiency trait (high root-shoot ratio), this did not lead to high P efficiency of the genotype since it had low P utilization efficiency (Chapter 2 and 3). The results revealed that both net photosynthetic and leaf dark respiration rates could not explain the lower NAR and hence lower P utilization efficiency observed with this genotype. However, considering the larger dry matter proportion allocated to the root and

higher P uptake and transport resulting in two-fold higher P concentration in plant dry matter of this genotype, it may be speculated that this genotype might have lost more carbon through root respiration, which is supported by results of Nielsen *et al*. (2001) who observed considerable carbon loss (amounting to 40% net carbon fixed by photosynthesis) through root respiration under P deficiency with P-inefficient common bean genotype. The higher carbon loss of genotype CGN 18233 might also be via root exudation as observed by Bekku *et al*. (1997) for different plant species.

Genotypes of crop species may use different P efficiency mechanisms (either uptake efficiency or utilization efficiency or both). Results of previous studies summarized in Table 1 show that uptake efficiency and traits associated with it has been observed with genotypes of several crop plants. However, studies focusing on mechanism of P utilization efficiency and information on traits related to P utilization efficiency are scarce.

Phosphorus efficiency in genotypes of barley, cabbage, cowpea, maize, rape, soybean, tea and wheat was mainly related to uptake efficiency. In *Brassica* cultivars, P utilization efficiency was observed whereas in common bean and rice genotypes, both uptake and utilization efficiency was reported.

With regard to improved root morphological traits, increased root hair length was for instance an important trait in improving the P uptake efficiency of barley, cabbage, cowpea and wheat genotypes. Other root morphological traits such as high root-shoot ratio and root length density at topsoil layer were also significant in improving P uptake efficiency of cowpea, maize, soybean, wheat and common bean genotypes. Furthermore, P-efficient genotypes of barley, cowpea, maize, rape, soybean, tea and wheat were also able to mobilize P via exudation of organic anions and/or acid phosphatases.

Common bean and rice genotypes demonstrated both P uptake and utilization efficiency mechanisms. The high P uptake efficiency in common bean genotypes was related to increased root length density at the topsoil layer for better topsoil foraging. Similar to most other crop genotypes, P-efficient

genotypes of common bean and rice were also able to mobilize P through exudation of either organic anions and/ or acid phosphatases. The high P utilization efficiency of common bean genotype under P limiting condition was related to efficient carbon budgeting (low carbon loss through root respiration) while that of rice genotype was due to high phostosynthetic rate. Similar to the common bean and rice genotypes, the potato genotypes investigated in the current study also showed both P uptake and utilization efficiency mechanisms.

Table 1: Mechanisms and traits associated with P efficiency of genotypes in some crop plants

P efficiency mechanism	Crop	P efficiency traits	References
Uptake efficiency	Barley	Longer root hair	Gahoonia *et al*., 1999 Gahoonia and Nielsen, 2004a
		P mobilization through exudation of organic anions, acid phosphatases and phytases	Asmar *et al*., 1995; Asmar, 1997; Gahoonia *et al*., 2000
	Cabbage	Longer root hair	Eticha and Schenk, 2001
	Cowpea	Longer root hair	Krasilnikoff *et al*., 2003
		High root-shoot ratio	
		P mobilization	
	Maize	High root-shoot ratio	Schenk and Barber, 1979
		P mobilization through exudation of organic anions & acid phosphatase	Gaume *et al*., 2001; Yun and Kaeppler, 2001; Singh and Pandey 2003; Liu *et al*., 2004; Corrales *et al*., 2007; Li *et al*., 2008
	Rape	P mobilization through release of protons and acid phosphatase	Zhang *et al*., 2009
	Soybean	High root-shoot ratio	Pan *et al*., 2008
		P mobilization through increased phosphatase activity	Ramesh *et al*., 2004
	Tea	P mobilization through proton release	Zoysa *et al*., 1999
	Wheat	Increased root hair length	Horst *et al*., 1993
		Increased root length density at topsoil layer	Egle *et al*., 1999; Manske *et al*., 2000
		P mobilization through exudation of acid phosphatase and phytase	Manske *et al*., 2000; Osborne and Rengel, 2002
Utilization efficiency	Brassica cultivars	Efficient re-translocation of P from metabolically inactive plant parts (older leaves & stems) to active parts (younger leaves)	Akhtar *et al*., 2006; Akhtar *et al*., 2008b
Both efficiency mechanisms	Common bean	Increased root length density at topsoil layer (U_pE)[1]	Liao and Yan, 2000; Miller *et al*., 2003
		P mobilization through exudation of organic anions (U_pE)[1]	Shen *et al*., 2002
		Lower carbon loss through root respiration (U_tE)[2]	Nielsen *et al*., 2001
	Rice	P mobilization through exudation of organic anions, acid phosphatase and proton release (U_pE)[1]	Ming *et al*., 2002
		High photosynthetic rate (U_tE)[2]	Yong-fu *et al*., 2001

1) U_pE= uptake efficiency 2) U_tE = Utilization efficiency

6. Crop genotypes efficiency mechanisms for other nutrients: the case of N and K

Similar to P efficiency, genotypes of crop species show an array of efficiency mechanisms and traits for other macronutrients such as nitrogen (N) and potassium (K). High N uptake efficiency of genotypes was related to increased root-length density in subsoil with maize (Wiesler and Horst, 1993,1994; Worku, 2005) and common bean (Kimani and Tongoona, 2008). With K, however, high uptake efficiency of genotypes was related to smaller root radius with rice (Jia *et al*., 2008) and both high root-shoot ratio as well as greater ability to mobilize non-exchangable K with potato and tomato (Chen and Gabelman, 2000; Trehan *et al*., 2005).

The high root-shoot ratio, smaller root radius and mobilizing ability, which were traits related to high K uptake efficiency were also reported as traits related to high P uptake efficiency (Table 1). High root-length density at subsoil was trait related to high N uptake efficiency. With P, however, the high root-length density at topsoil layer was related to high uptake efficiency as observed with common bean genotypes (Liao and Yan, 2000; Miller *et al*., 2003). This is because P, unlike N, is immobile in the soil and thus available P concentrates in the topsoil layer.

On the other hand, high N utilization efficiency in maize genotypes was related to delayed leaf senescence, higher leaf chlorophyll concentration, ability to retain activities of enzymes related to photosynthesis (higher photosynthetic efficiency) and ability to remobilize N from vegetative parts at grain filling stage (Hirel *et al*., 2001; Maranville and Madhavan, 2002; Paponov *et al*., 2005; Worku *et al*., 2007; Schulte auf'm Erley *et al*., 2007). With K, high utilization efficiency was related to greater ability to translocate K from non-photosynthesizing organs to leaves maintaining higher photosynthetic capacity at grain filling stage and also to greater ability to maintain PS-II photochemical efficiency as observed with rice genotypes (Yang *et al*., 2003, 2004; Jia *et al*., 2008).

Similar to N and K utilization efficiency mechanisms described above, crop genotypes also showed higher photosynthetic capacity as a mechanism of P utilization efficiency as observed with rice genotype (Yong-fu *et al*., 2006). Like for N and K, high P remobilization ability was also accounted for higher P utilization efficiency in *Brassica* cultivars (Akhtar *et al*., 2008b).

7. Outlook

From results of the present study it was suggested that the high P utilization efficiency of genotype CGN 17903 was due to high NAR, which was speculated to be due to lower root carbon cost. It is suggested that this speculation be further elucidated by investigating the genotypes for the carbon cost of root respiration and exudation.

Previous studies showed that there was enhanced P uptake due to mycorrhizal association under P limiting condition with potato, indicating that potato can benefit from the association (Davies *et al*., 2005a, b). Moreover, mycorrhizal colonization of 70-80% was observed with potato (Bhattarai and Mishra, 1984). However, since the efficiency of the association in enhancing growth and yield of potato depend on a suitable combination of cultivar and arbuscular mycorrhiza (AM) fungi specie used (Yao *et al*., 2002), evaluation of the genotypes for ability to benefit from inoculation with different mycorrhiza strains can be suggested for future work.

The useful traits related to P efficiency, in the present study, were identified for both cultivated and wild genotypes. In case of the wild genotype, the trait related to P efficiency need to be transferred to adapted P-inefficient cultivars through breeding. However, since P efficiency is a quantitative trait controlled by multiple genes (Li *et al*., 2005) the quantitative trait loci (QTLs) or major genes linked to P efficiency need to be identified as a first step of the breeding work. It was reported that quantitative traits related to P efficiency in common bean have been successfully transferred from P-efficient cultivar into agriculturally adapted inefficient cultivar (Schettini *et al*., 1987). Thus following the detection of QTLs or major genes related to P efficiency, the useful

quantitative traits related to P efficiency in the wild potato genotype need to be incorporated into adapted potato cultivars lacking these traits through breeding.

SUMMARY

Potato has a high phosphorus (P) fertilizer requirement for optimum growth and yield. However, because of low P availability in nearly 67% of the cultivated soils, P deficiency is a major factor constraining crop production. To avert this problem, the use of genotypes/ cultivars with high P efficiency is an option for sustainable potato production in inherently low P soils as well as in soils with high P fixing capacity.

This study was conducted with the objectives to screen potato genotypes for P efficiency, to elucidate the mechanism of P efficiency and to evaluate the contribution of root traits to P uptake of the genotypes using mechanistic simulation model.

To achieve these objectives, plants were grown in controlled growth chamber in soil at two P levels (low and high) and in nutrient solution under three P regimes (low, medium and high). In screening experiments, the genotypes were grown in soil at two P levels and evaluated for P efficiency in terms of high shoot dry matter yield (SDMY) and relative shoot growth rate (RGR_s) under P limiting condition. Four genotypes, two of which were consistently P-efficient and the other two, which were consistently P-inefficient in terms of both SDMY and RGR_s at low P were selected for investigation of P efficiency mechanisms. These four genotypes with contrasting P efficiency were further evaluated in soil experiments and additional plant parameters related to P efficiency such as shoot P concentration, root length and root hair length were determined. The influence of morphological root characteristics was evaluated using mechanistic simulation model. To investigate the mechanism of P utilization efficiency of the genotypes, a nutrient solution experiment was conducted and data for plant leaf area, gas exchange rate and leaf starch content were determined.

The results showed that

(1). Both genotypes CGN 17903 and CIP 384321.3 had higher SDMY and RGR_s (P-efficient) compared to genotypes CGN 22367 and CGN 18233 (P-

inefficient) at low P supply. The P efficiency of genotype CGN 17903 was exclusively related to high P utilization efficiency whereas for genotype CIP 384321.3 it was related to both high uptake efficiency and intermediate P utilization efficiency.

(2). The higher P utilization efficiency of genotype CGN 17903 (higher relative plant growth rate (RGR_p) at low P) was caused by higher net assimilation rate (NAR).

(3). The high P uptake efficiency of genotype CIP 384321.3 was due to higher root-shoot ratio.

(4). The P inefficiency of genotype CGN 18233 was related to low P utilization efficiency despite its high uptake efficiency in terms of higher root-shoot ratio. This indicates that besides P uptake efficiency, P utilization efficiency also determines P efficiency of a genotype. The low P utilization efficiency of this genotype was related to low NAR and this low NAR was speculated to be due to high carbon loss through root respiration and/or exudation.

(5). The P inefficiency of genotype CGN 22367 was related to both low P uptake efficiency and intermediate P utilization efficiency. The low P uptake efficiency of this genotype was related to low root-shoot ratio whereas the intermediate P utilization efficiency might be due to higher leaf dark respiration.

(6). Although the P-efficient genotypes had longer root hairs this did not differently influence the P uptake rate as well as the width of the P depletion zone around the root surface of P-efficient genotypes compared to that of the P-inefficient genotypes.

(7). Since the predicted and observed P uptake agreed fairly well indicating that the major processes involved in P transport and root characteristics of the genotypes affecting P uptake were well described and also since there was no difference in P uptake rate between the genotypes at low P level it is suggested that P mobilization was not involved.

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II. EDUCATION

Year	**School/University**	**academic degree**
Tertiary education		
Oct. 2005-upto date	PhD student at Leibniz University of Hannover, Faculty of Natural Science, Germany	PhD in Horticulture
Sep. 2000-Jun. 2002	MSc student at Indian Agricultural Research Institute, New Delhi, India	MSc in Horticulture
Sept.1991-Jul. 1995	BSc student at Alemaya University of Agriculture, Ethiopia	BSc in Plant Sciences
Secondary education		
Sep.1986- May1991	Ambo Comprehensive Senior Secondary School, Ethiopia	Ethiopian School Leaving Certificate
Primary education		
Sep.1978- Jun. 1986	Ejaji Junior Secondary School, Ethiopia	Certificate

Special course

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III. WORK EXPERIENCE

Sep. 1995-Aug. 1998	Junior researcher (cereal crop improvement) at Bako Research Center, Ethiopia
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IV. PUBLICATIONS

Balemi, T. and Schenk, M.K. (2009): Genotypic difference of potato in carbon budgeting as a mechanism of phosphorus utilization efficiency. *Plant and Soil* (DOI: 10.1007/s11104-009-9897-0).

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