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The effect of maternal environment on seed and seedling quality in tomato

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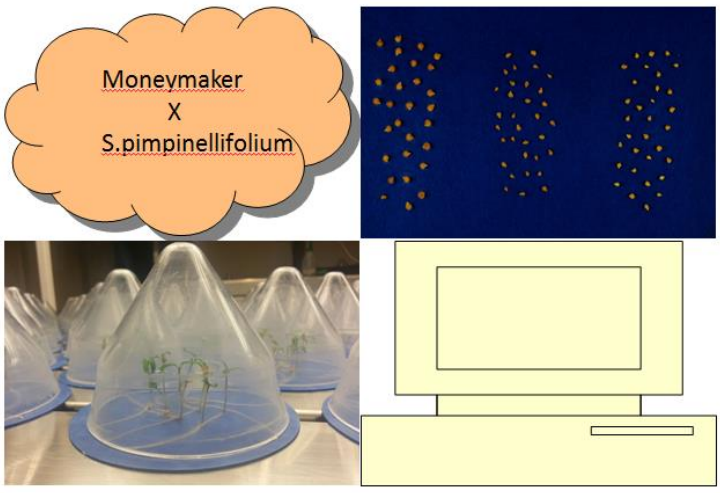

Supervised by: **Wilco Ligterink , Shuangchen Chen & Nafiseh Geshnizjani**

Table of contents

Contents

Abstract

Seed quality is a complex term which depends on genetic and environmental factors. Growth and development of plant not only effected by existing environment condition. It also depends on parental environmental condition such as light, temperature during seed development and maturation stage. We have conducted factorial experiment to know effect on seed and seedling quality in tomato on seed developed under different nitrate and phosphate treatment. On seed developed under different nitrate and phosphate condition of tomato RIL population and parents, we have measured the germination percentage, fresh root weight, dry root weight, fresh shoot weight and dry shoot weight. We have found that effect of nitrogen and phosphorous on seed quality is genotype dependant. For instance in line 261 seed developed under very high nitrate showed less germination on other hand in line 221, 230 and money showed higher germination in seed developed under high nitrate condition. In line 254, 261, 290, money and pimp no difference in different phosphate treatment with respect to fresh shoot weight. However fresh shoot weight found low in seed developed under very low phosphate condition in line 205, 221 and 230 compare to other phosphate condition.

Introduction

Seed quality is a complex term which depends on genetic and environmental factors[1]. During the seed developmental stage, the seed quality is governed by the genetic and environment interactions [1, 2]. Seed germination ability, dormancy, longevity, seedling vigour and uniform growth are the important elements of seed quality [1]. Several definitions of seed quality are given in literature, for instance the capability of a seed to germinate in different environmental conditions and grow into healthy plantlets[3], seed quality is the viability and vigour elements of seeds which allows it to develop into healthy

seedlings under various environmental conditions [4]. In practical terms, the definition of seed quality also depends on the type of industry. In food industry, seed quality may depend on high starch and oil content. In agriculture, seed quality depends on germination and seedling establishment in various environmental conditions [1].

Seed companies focus on improving seed quality at harvest and postharvest stages by controlling harvesting time and postharvest treatments such as sorting, grading and priming etc. However, seed quality can also be improved by controlling production environments[1]. As an example in studies on *Arabidopsis thaliana*, Elwell et al (2011) found that progeny shows differences in seed weight, seed germination and early seedling development stage when the same genotype is grown in different environments [5][5] . Growth and development of plant not only effected by existing environment condition[5]. It also depends on parental environmental condition such as light, temperature during seed development and maturation stage [6]. Various studies show the impact of production environments on seed quality elements as described below.

For the effect of temperature in *Arabidopsis thaliana*, Blonder et al. (2007) found that seeds from parents grown at warm temperature (25˚C) showed quicker germination and root elongation than seeds from parents grown under cold conditions (15˚C) [7]. Similar results were shown in a study of *P. Lanceolata* [8]. In an experiment conducted by Demir et al (2004) in watermelon (Citrullus lanatus) dry mass and longevity of seeds was bigger in cooler growing sites compared to warmer growing sites. In another experiment conducted by [9] with tomato (*Solanum lycopersicum*), seedling establishment from seed harvested in spring was higher than in autumn [10].

In tomato, long day conditions during fruit ripening, results in seeds with less germination than seeds of maternal plants exposed to shorter day length [11].

Limited water availability during seed development had little effect on seed quality of *Lactuca sativa* L. Although, there was an increase in seed weight but there were fewer seed production per plant [2]. On the other hand, no significant effect was found on germination and primary seedling growth in tomato from seed developed under different irrigation interval [12] .

For nutrient effect in *Solanum lycopersicum,* higher dose of nitrogen application improve germination of progeny [13]. Higher dose of nitrogen and phosphorous in combination to mother plant can help to improve the germination rate and seedling growth of progeny [14]. In experimental study on *Abutilon theophrasti* by Wulff et al 1992, they have found that the increase in maternal nutrient supply has positive effect on early stage of seedling growth of progeny [6]. However, the maternal nutrient effect also depends on genotype and progeny growth environmental condition[6]. On other hand, in the field experiment conducted by Singh and Kumar 2010 on effect of phosphorus and nitrogen fertilization on seed production of tomato, they have found that with increase amount of nitrogen application showed positive effect on seed yield and higher phosphorus application show negative effect on seed yield. However, no significant effect of nitrogen and phosphorus was found with respect to the seed quality [15]. The germination percentage in *Synapsis arvensis* was found to be decreased from seed progeny of parent grown under high nitrogen percentage [16]. High amount of nutrient and day light to mother plant in *Campanula americana* resulted in the high seed weight of the progenies but low germination percentage [17]. Seeds of *Amaranthus retroflexus* grown under high nitrogen level showed dormancy while that on

low nitrogen level showed higher germination rate. However, low nitrogen and gibberellic acid application during seed development stage showed highest germination percentage [18].

Different environmental factors affect the seed and seedling quality in various crops as discussed above. The aim of this study is to examine the effect of nitrogen and phosphorous on seed and seedling quality in tomato. In earlier experimental studies only one or two genotypes were studied. In this study, we used lines from a recombinant inbred line (RIL) population of *Solanum lycopersicum cv.* Moneymaker and *Solanum pimpinellifolium* and the parents. *Solanum lycopersicum* is a cultivated tomato species and seed germination and early seedling growth is sensitive to abiotic stress. *Solanum pimpinellifolium* is a wild tomato species with more tolerance to abiotic stress during seed germination and early seedling growth [19]. The use of this genetic variation in our study allows us to not only see the effect of the maternal environment on seed and seedling quality, but also if there is genotype by environmental interaction for these traits.

Material and methods

Plant material

Seed batches from the recombinant inbred line (RIL) population of *Solanum lycopersicum cv.* Moneymaker and *Solanum pimpinellifolium* and the parents are used in our experiments. The RIL lines were 205, 221, 230, 254, 261 and 290. The RIL lines and parents were grown under different phosphate and nitrate concentration during flowering and seed development (Table 1).

Table 1: The different nitrate and phosphate concentrations during growth of the RIL lines the parents of the RILs.

	Nitrate	Phosphate
Control	14mM	1000µM
Very low nitrate	0.2mM	1000µM
Low nitrate	3mM	1000µM
High nitrate	20mM	1000µM
Very high nitrate	36mM	1000µM
Very low phosphate	14mM	30µM
Low phosphate	14mM	100µM
High phosphate	14mM	5000µM
Very high phosphate	14mM	10000µM

Germination assay

The seeds were sown in transparent plastic trays (from DBP plastics, size 15 X 21cm) for germination. Each germination tray had 2 blue filter papers (Blue blotter paper, Anchor paper company, size 5.6 X 8 inches) and 50ml -0.5MPa NaCl. In each tray 3 sample of around 25 to 35 seeds were sown by using a mask (Fig. 1). A randomized complete block design was used and 12 trays were piled up and on the top and the bottom of each pile, one tray containing 2 white filter papers and 50ml MQ water were placed and the top was covered with a lid to avoid unequal evaporation. Then pile was put in a transparent plastic bag and placed at 4˚C for 72 hours for stratification. Afterwards the bags were transferred to an incubator at 25˚C in light for germination. The trays were kept in the incubator for 10 days and during the initial 3 days pictures were taken of each tray twice a day. From the 4th to 10th day pictures were taken once a day. Pictures were taken with a Nikon D80 camera

fixed to a Kaiser Stand at 60cm distance. The camera was connected to a computer via Nikon camera control pro software version 2.

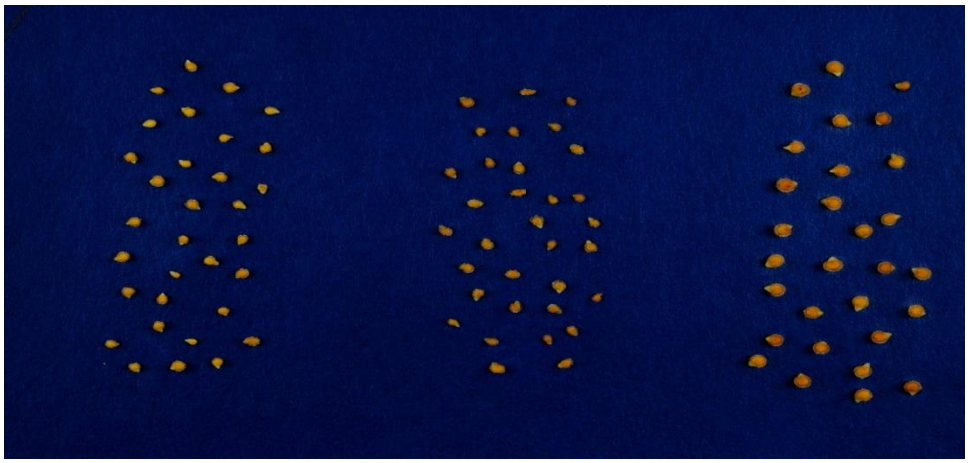

Figure 1. The arrangement of three different seeds samples sown in a germination tray

Seedling growth

Seedling growth was measured by the method described by Khan et al (2012) [4]. In short, seedling growth was measured by performing a germination assay as above till time point 50 percent viable seed germination on MQ instead of NaCl. The first 10 germinated seeds were discarded per sample. From the remaining germinated seeds 10 seeds per sample were transferred to a Copenhagen table at 22.5 ˚C. The transferred seeds were allowed to grow for 10 days after which fresh weight of root and shoot of each sample. Subsequently the shoots and roots were dried in an oven at 80˚C for 48 hours to obtain dry root and shoot weight.

Data analysis

Germination traits were measured by using the germinator package as described in Joosen et. al 2010 [20] and in a more detailed manual from the WSL. Different parameters were obtained. In this report the three parameters used are t10 (time to germinate first 10% of viable seed), t50 (time to germinate 50% viable seed) and gMAX (maximum seed

germination percentage) to analyse the effect on seed germination. In genotype if germination percentage remain below 10% that genotype excluded from t10 statistical analysis. Also in t50 statistical analysis if germination percentage remain below 50% that genotype excluded.

To analyse seedling trait as fresh and dry shoot weight and fresh and dry root weight one way ANOVA analysis done in Genstat 16sp1 edition was used. To check significant effects of each treatment a fischers least significant difference test (LSD) was used at a=0.05.

Results

To see the effect of nitrate and phosphate treatment during seed development on progeny germination we have measured the progeny germination percentage, t_{50} and t_{10}. The same seeds were used to study the effect of nitrate and phosphate treatment during seed development on progeny seedling growth. In this respect we have measured fresh root and shoot weight and dry root and shoot weight of 10 days old seedlings.

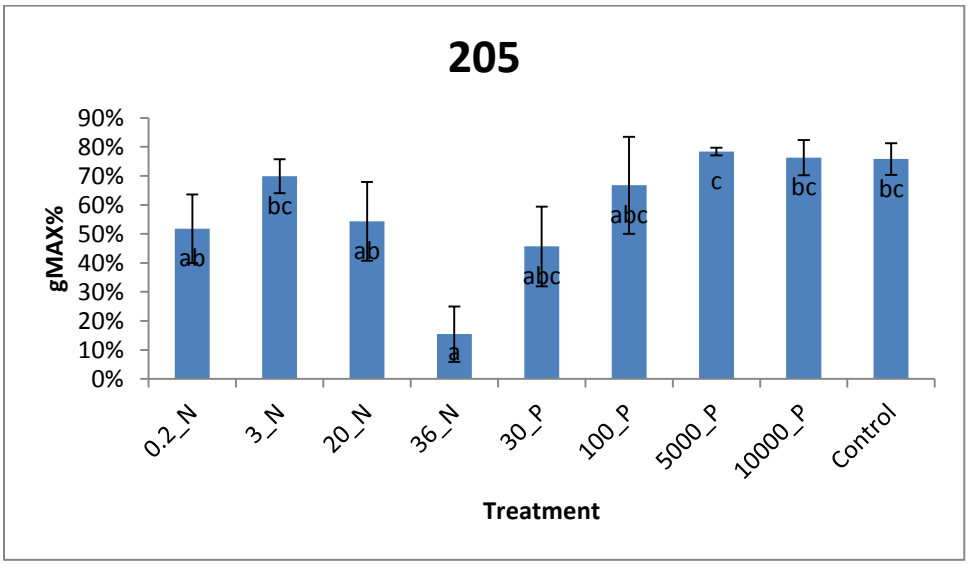

Figure 2 Germination percentage (gMAX%) and SE ± in line 205 after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05.

In line 205 much lower germination is found in the seeds of mother plants treated with very high nitrate condition (15%) compare to seeds of control (76%), low nitrate (70%), high phosphate (78%) and very high phosphate (76%) treated mother plants. Among other treatments in relation to control no significant difference in germination percentage was observed. However the germination percentage was more in seeds of high phosphate (78%) treated mother plants than seeds of very low nitrate (52%) and high nitrate (54%) treated mother plants (Fig.2).

For line 205 we have not observed significant differences between different nitrate and phosphate concentrations on the t_{50} (Data not shown).

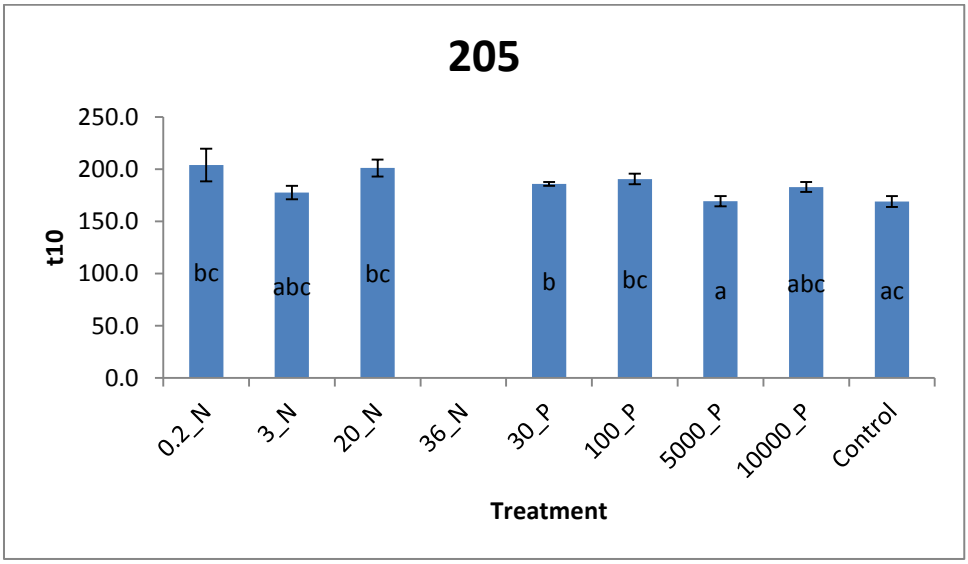

Figure 3. Time in hours and SE ± for 10 percent viable seeds to germinate. In line 205 after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicates statistical significant difference by student t test; α=0.05.

For the t_{10}we have only observed significant difference between seeds of very low phosphate treated mother plant and seeds of control and high phosphate treated mother plant. The seeds of very low phosphate treated mother plant took 185.7 hours, seeds of control 168.9 hours and seeds of high phosphate 169.4 hours to reach t_{10} (Fig.3).

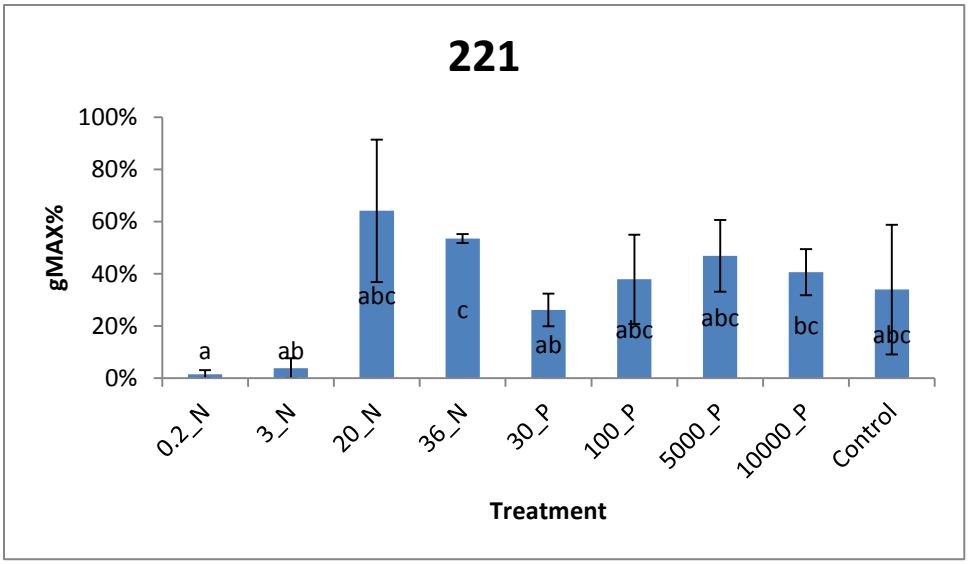

Figure 4. Germination percentage (gMAX%) and SE± in line 221 after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05.

In line 221 the seed germination percentage in seeds of very high nitrate mother plant (53%) was higher than seeds of very low (2%), low nitrate (4%) and very low phosphate (26%) treated mother plant. However with respect to control we have not found any significant effect of phosphate and nitrate on seed germination percentage (Fig.4).

For the t_{10} we have not observed any significant differences in phosphate and nitrate treatment of the mother plant (Data not shown). Values for t_{50} were not calculated for line 221 due to too low germination percentages.

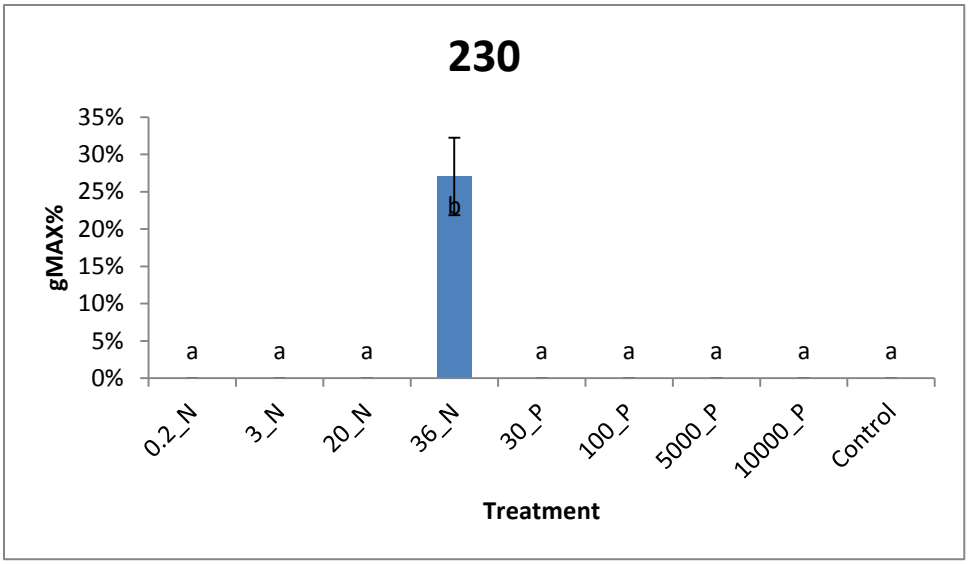

Figure 5. The germination percentage (gMAX%) and SE ± in line 230 after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05.

In line 230 we have observed the germination in seeds of very high nitrate treated plant which was 27% only. In other treatment we have not observed any germination (Fig. 5). Therefore, Line 230 is excluded from t_{50} and t_{10} analysis.

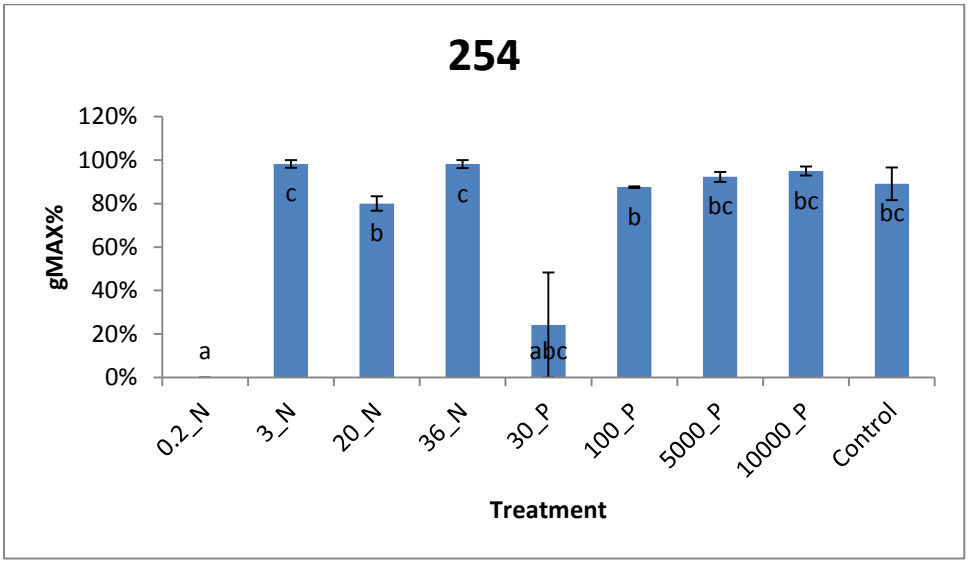

Figure 6. The germination percentage (gMAX%) and SE ± in line 254 after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05.

In line 254 the 98% germination in seeds of very high and low nitrate treated mother plant observed. It was higher compare to seeds of high nitrate and low phosphate treated mother

plant. In seeds of high nitrate treated mother plant 80% and in seeds of low phosphate treated mother plant 88% germination observed (Fig.6).

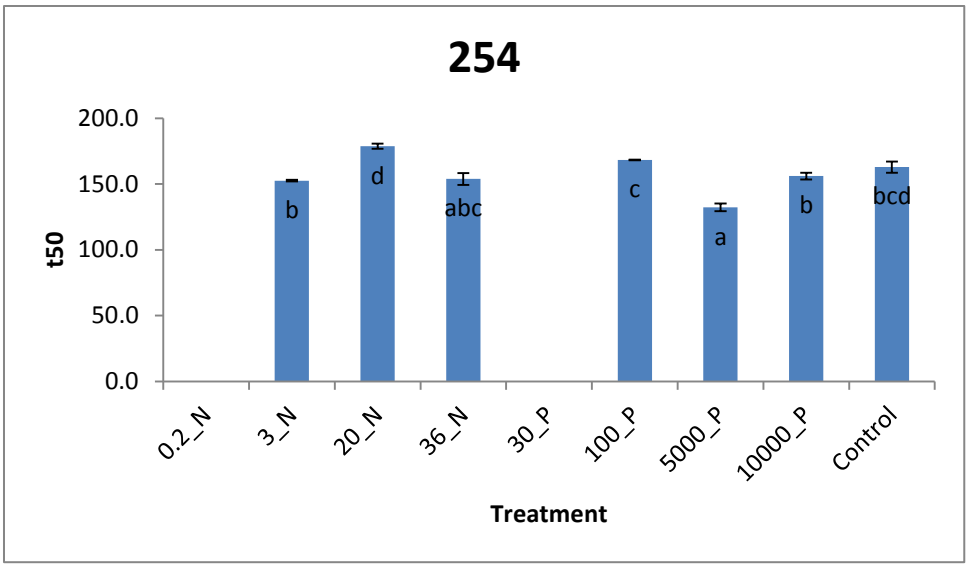

Figure 7. Time in hours and SE ± for 50 percent viable seeds to germinate. In line 254 after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05.

For thet_{50} analysis we have not observed significant differences with respect to control except seeds of high phosphate treated mother plant. However we have observed the significant differences for different phosphate and nitrate treatment. Reach to t_{50} seeds of high phosphate took 132 hours, very high nitrate took153 hours and low nitrate took 152 hours, low phosphate took 168 hours and high nitrate took 178 hours in order observed (Fig.7).

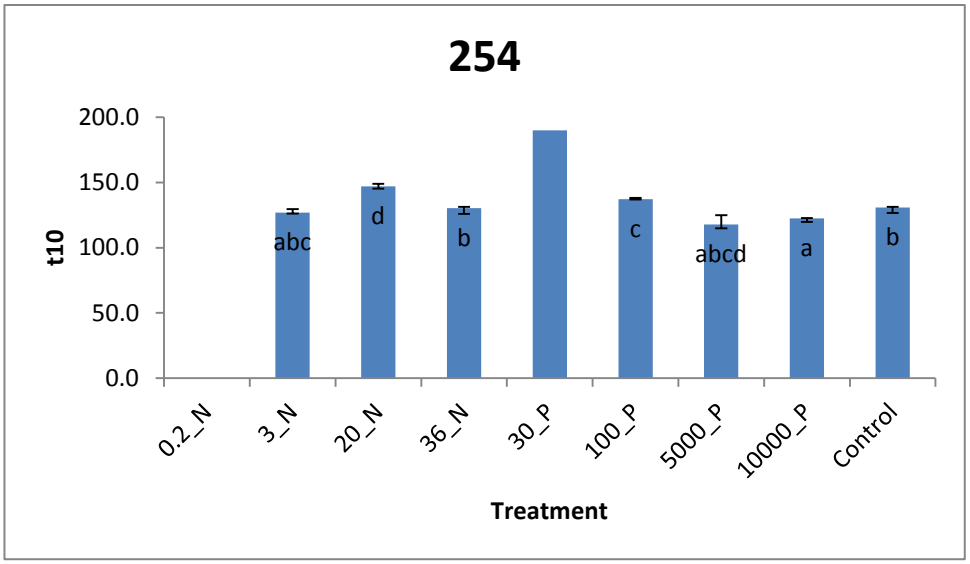

Figure 8. Time in hours and SE ± for 10 percent viable seeds to germinate. In line 254 after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05.

For the t_{10} with respect to control (130.9hr) seeds of very high phosphate (122.4hr) treated mother plant germinated early. On other hand seeds of low phosphate (137.2hr) treated mother plant and seeds of high nitrate (147hr) treated mother plant germinated later (Fig.8).

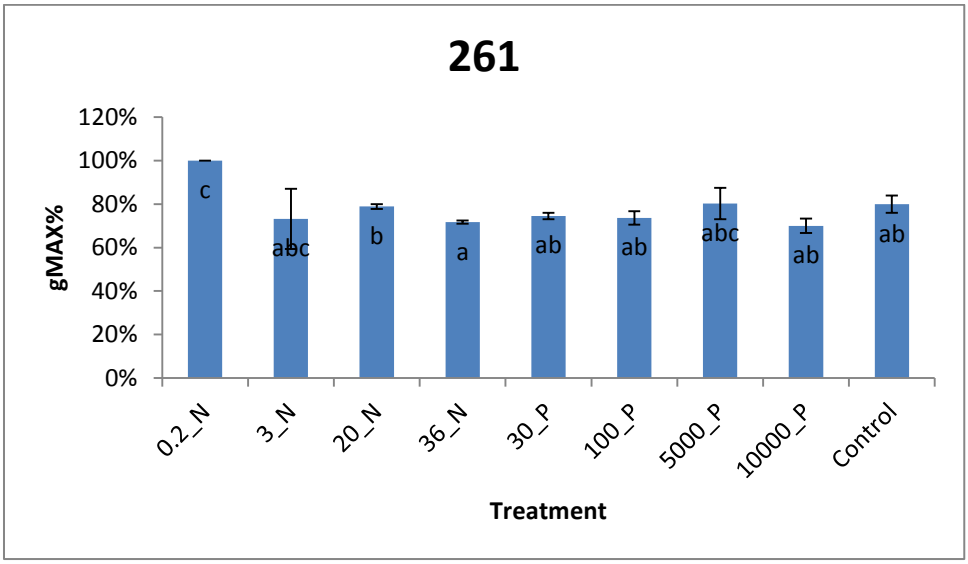

Figure 9. The germination percentage (gMAX%) and SE ± in line 261 after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05.

In line 261except seeds of very low nitrate treated mother plant with respect to control no significant deference observed. In line 261 the germination was maximum with seeds of

mother plant low nitrate treated (100%). However not significant difference from seeds of low nitrate treated mother plant and seeds of high phosphate treated mother plant (Fig.9).

In line 261 in t_{50} analysis we have not observed any significant differences. Data not shown.

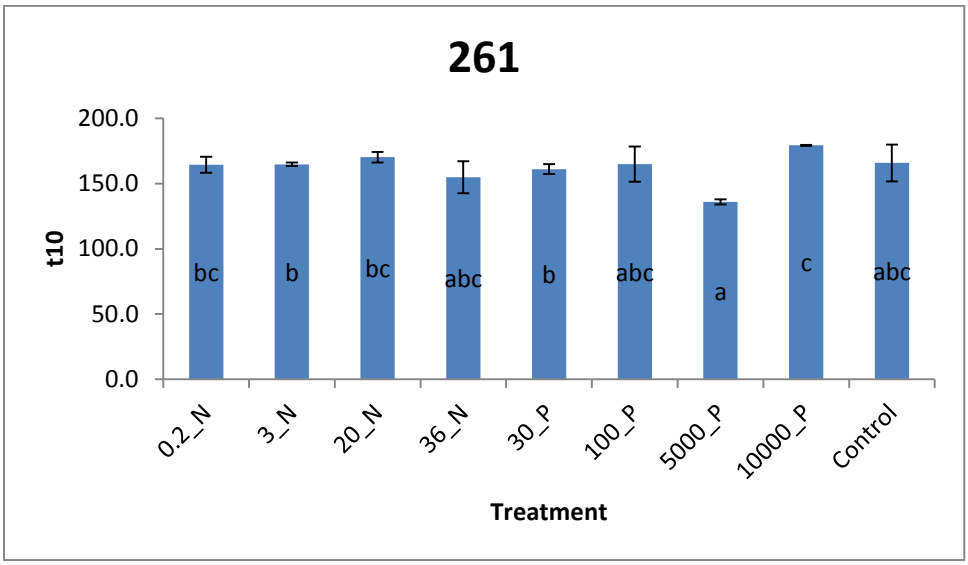

Figure 10 Time in hours and SE ± for 10 percent viable seeds to germinate. In line 261 after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05.

For the t_{10} analysis with respect to control no significant differences we have observed. However seeds of mother plant with high phosphate treated (136hr) reached early than seeds of very low phosphate treated mother plant (161hr), seeds of low nitrate treated mother plant (164.7hr), seeds of very low nitrate treated mother plant (164.3hr) and seeds of high nitrate treated mother plant (170.2hr) (Fig.10).

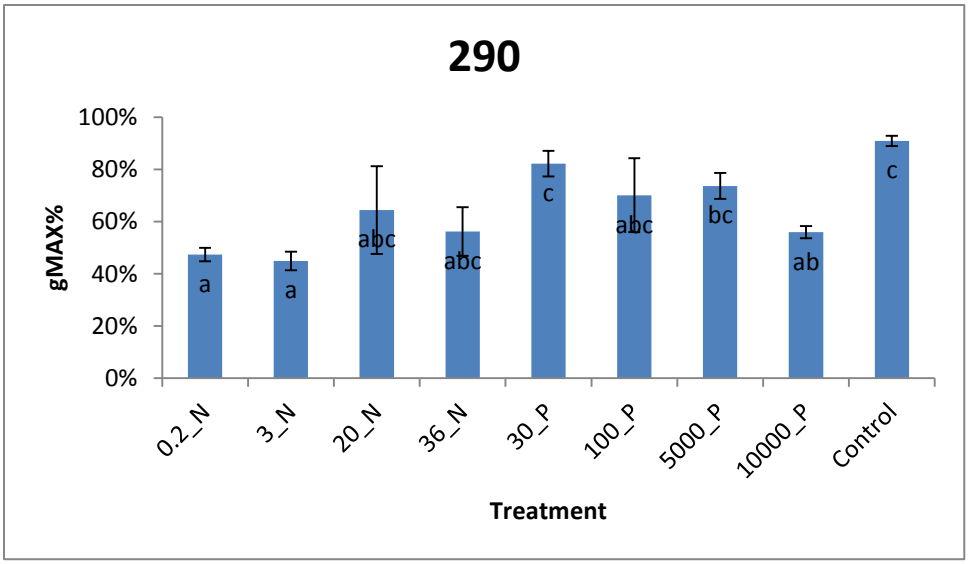

Figure 11 The germination percentage (gMAX%) and SE ± in line 290 after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05.

In line 290 with respect to control (91%) and seeds of very low phosphate treated mother plant (82%) significantly less germination observed in seeds of very high phosphate treated mother plant (56%), seeds of low (45%) and very low nitrate treated mother plant (47%). The effect of low phosphate, high nitrate and very high nitrate not significantly different from others. (Fig.11).

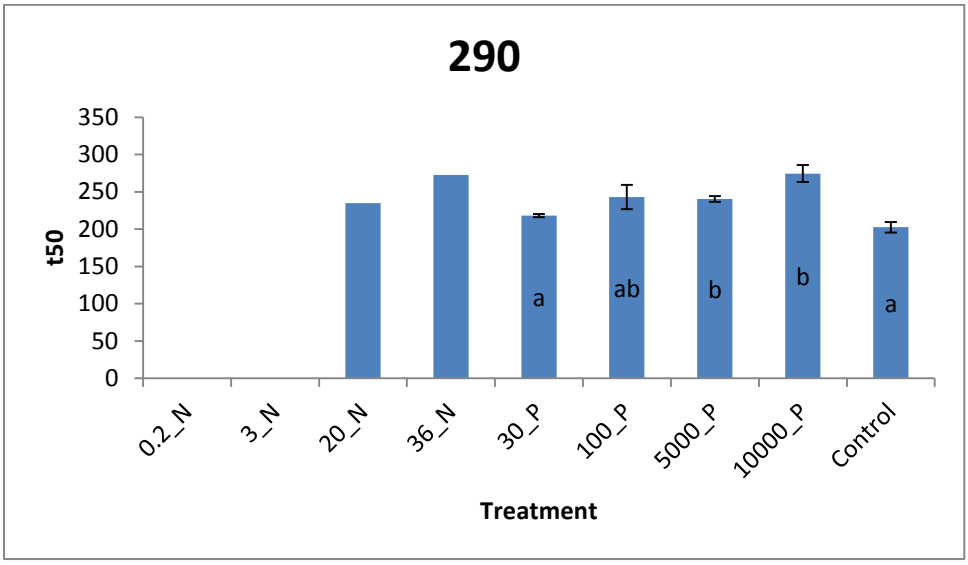

Figure 12. Time in hours and SE ± for 50 percent viable seeds to germinate. In line 290 after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05.

For the t_{50} analysis the control (202.5hr) and seeds of very low phosphate treated mother plant (218.1hr) faster compare to seeds of high phosphate treated mother plant (240.6hr) and very high phosphate treated mother plant (274.5hr) (Fig.12).

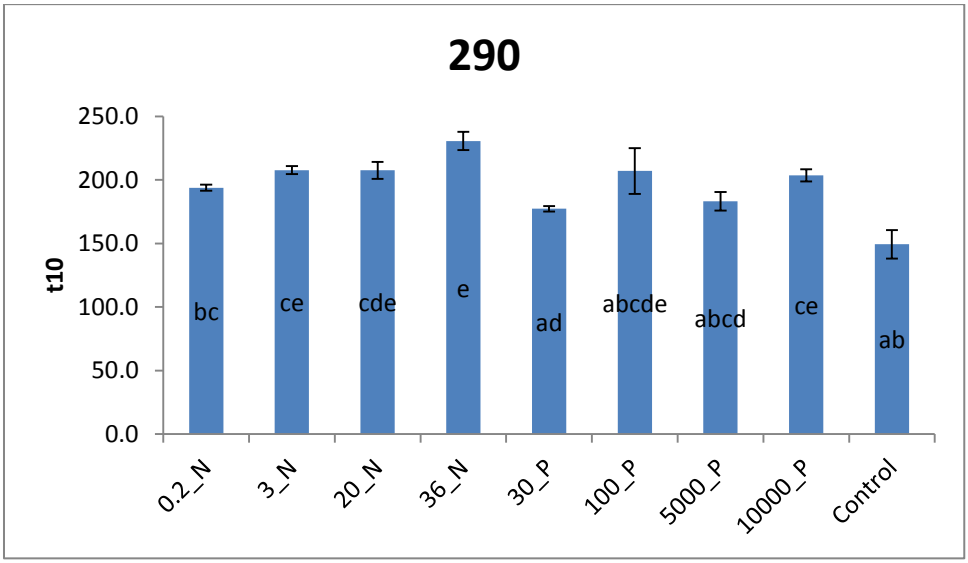

Figure 13. Time in hours and SE ± for 10 percent viable seeds to germinate. In line 290 after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05.

For the t_{10} analysis seeds of control (149.3hr) was faster than seeds of low nitrate (207.7hr), high nitrate (207.5hr) and very high nitrate (230.6hr) treated mother plant and seeds of very high phosphate treated mother plant (203.6hr). The seeds of very high nitrate treated mother plant (230.6hr) delayed than seeds of mother plant treated with very low nitrate (149.3hr) and very low phosphate treated mother plant (177.3hr) (Fig.13).

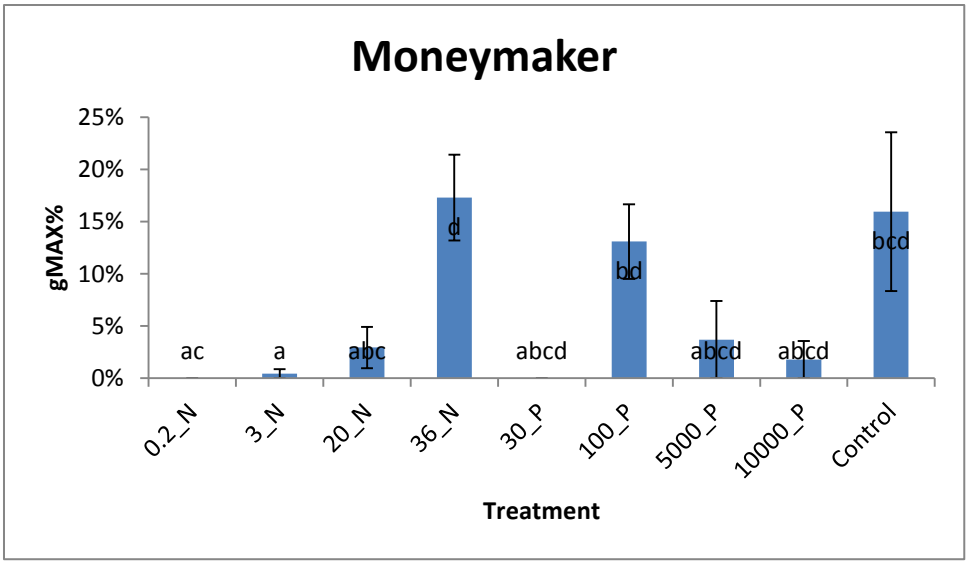

Figure 14 The germination percentage (gMAX%) and SE ± in line money after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05.

In money seeds of very high nitrate (17%) and low phosphate (13%) treated mother plant higher germination percentage observed than seeds of very low nitrate (0%) and low nitrate (0%) treated mother plant. From t_{50} and t_{10} analysis money is excluded (Fig.14).

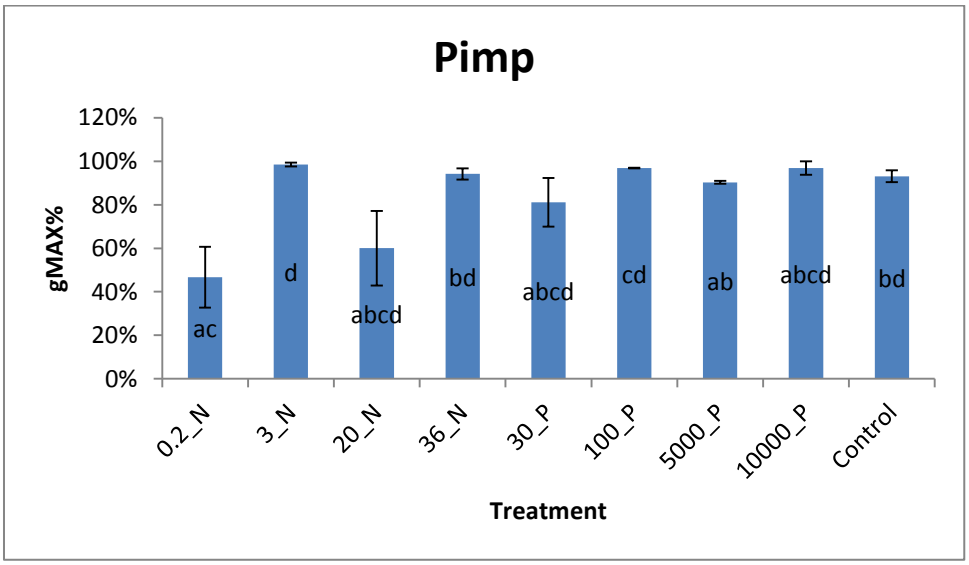

Figure 15 The germination percentage (gMAX%) and SE ± in line pimp after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05.

In line pimp the germination percentage observed low in seeds of very low nitrate treated mother plant (47%) compare to seeds of very high nitrate treated mother plant (98%) and control (93%). No significant different effect of other treatment observed (Fig.17).

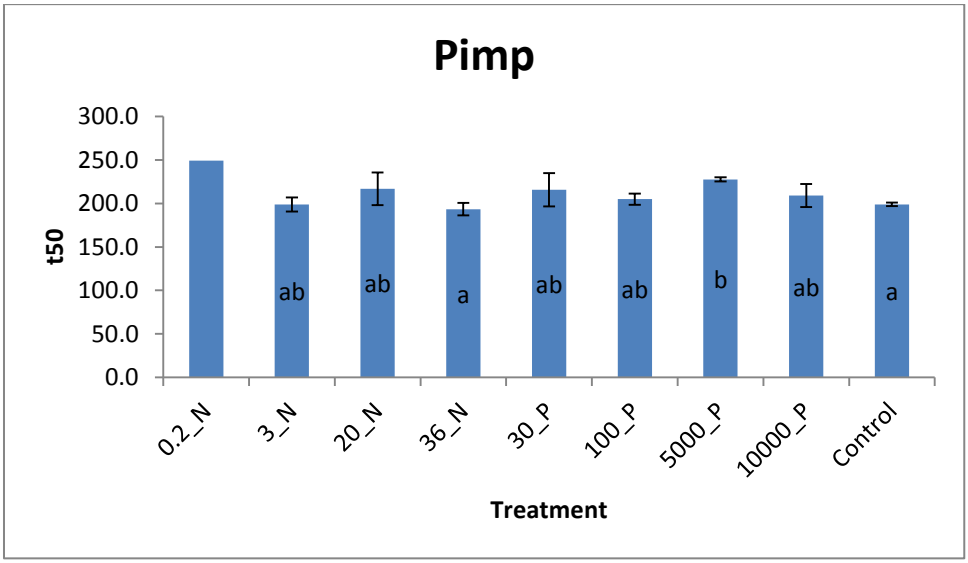

Figure 16 Time in hours and SE ± for 50 percent viable seeds to germinate. In line pimp after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05

For the t_{50} analysis seeds of control (198.9hr) and seeds of very high nitrate plant (193.3) germinated faster than seeds of high phosphate treated mother plant (227.6hr). No significant differences observed for other treatment effect (Fig.16).

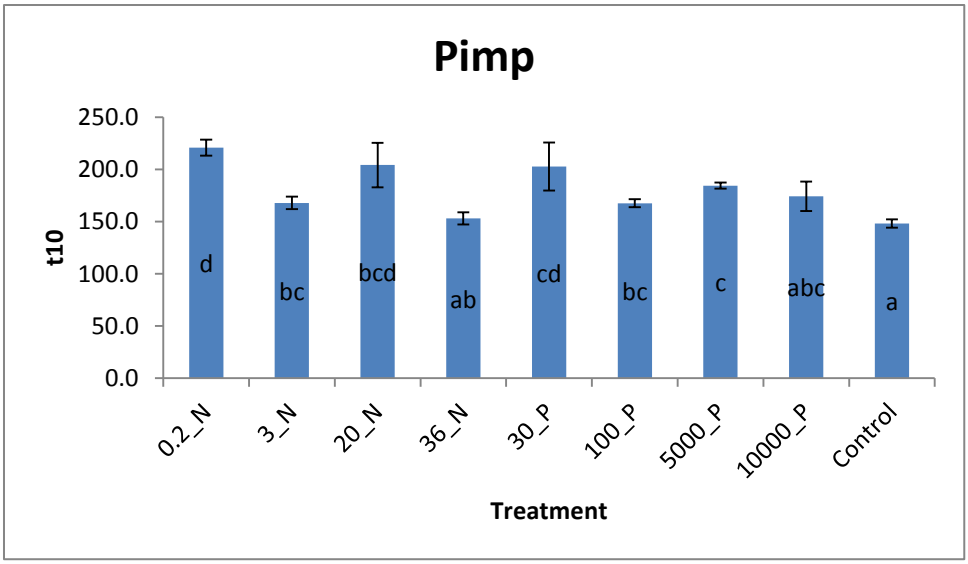

Figure 17 Time in hours and SE ± for 10 percent viable seeds to germinate. In line pimp after 10 days incubation at 25˚C in -0.5MPa salt concentration. Different letters indicate statistical significant difference by student t test; α=0.05

For the pimp in t_{10} analysis seeds of control (148hr) consumed less time than seeds of low phosphate (167.6hr), high phosphate (184.5hr) and very low phosphate (202.7hr) treated mother plant and seeds of very low nitrate (220.7hr) treated mother plant. Seeds of very low nitrate (220.7hr) treated mother plant required more time than seeds of low nitrate (167.9hr) and very high nitrate (153.1hr) treated mother plant and seeds of low phosphate (167.6hr), high phosphate (184.5hr) and very high phosphate (174.3hr) treated mother plant. The seeds of high phosphate (184.5hr.) treated mother plant consumed more time than seeds of very high nitrate treated mother plant (174.3hr) (Fig.17).

Result on seedling quality traits fresh root and shoot weight and dry root and shoot weight after 10days growth on Copenhagen table in line 205, 221, 230, 254, 261, 290, money and pimp.

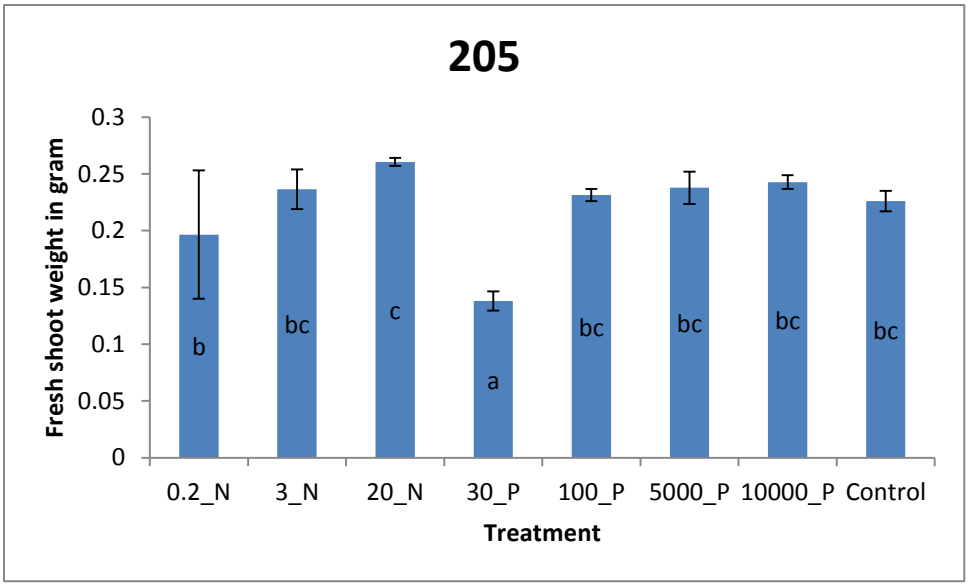

Figure 18 The Means and SE± of fresh shoot weight in gram 10 days after growth on copenhagen table in line 205. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05

In line 205 with respect to fresh shoot weight only in seeds of mother plant treated under very low phosphate low shoot weight observed compare to all other nitrate and phosphate treatment. Furthermore fresh shoot weight of seed of mother plant treated under high nitrate observed more than seeds of mother plant treated under very low nitrate condition (Fig 18).

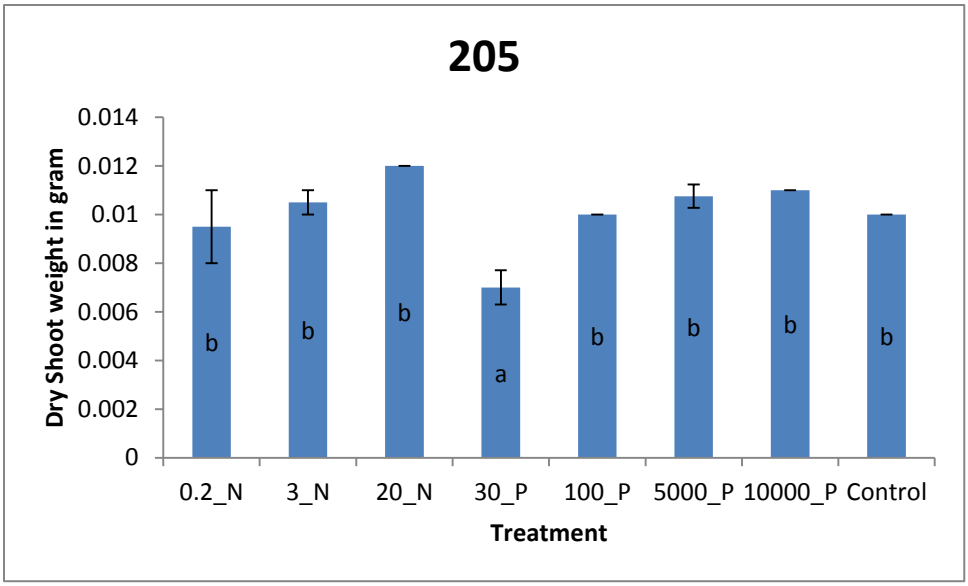

Figure 19 The Means and SE± of dry shoot weight in gram 10 days after growth on copenhagen table in line 205. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05.

In line 205 for the dry shoot weight no significant difference found in different treatment except seeds of very low phosphate treated mother plant. The dry shoot weight from seeds of very low phosphate treated mother plant found low compare to all other phosphate and nitrate treatment effects (Fig 19).

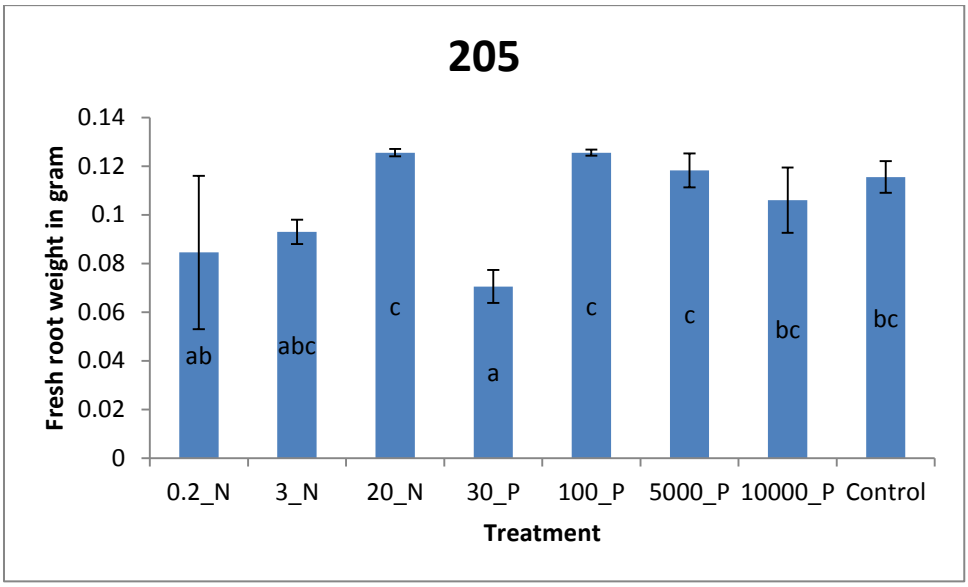

Figure 20 The Means and SE± of fresh root weight in gram 10 days after growth on copenhagen table in line 205. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05

In line 205 for the fresh root weight from seeds of very low phosphate treated mother plant observed low compare to seeds of low phosphate, high phosphate and very high phosphate and high nitrate and control. The fresh root weight from seeds of high nitrate treated mother plant found higher than seeds of very low nitrate treated mother plant (Fig 20). In line 205 with respect to dry root weight no significant difference observed. Data not shown..

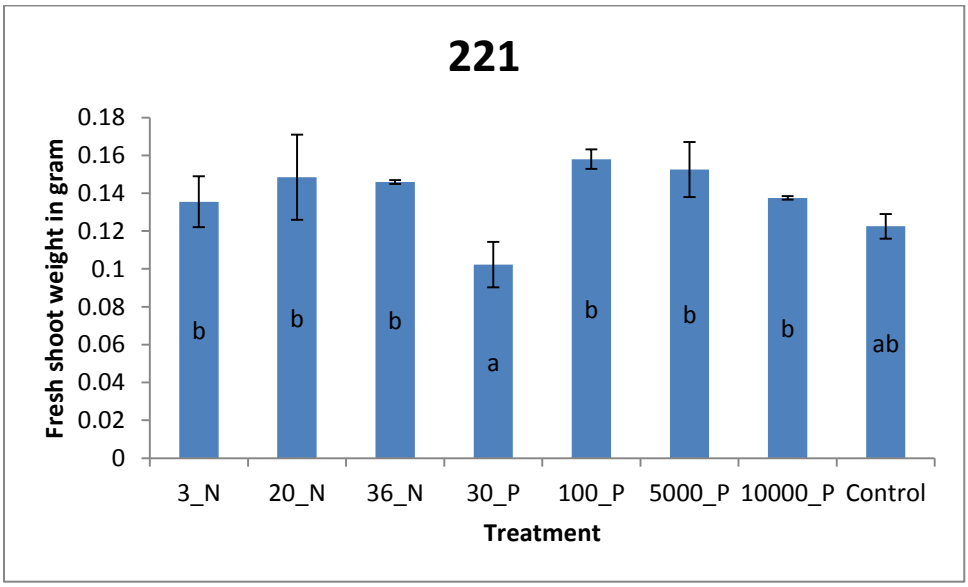

Figure 21 The Means and SE± of fresh shoot weight in gram 10 days after growth on copenhagen table in line 221. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05.

In line 221 for the fresh shoot weight no significant difference found in different treatment except seeds of very low phosphate treated mother plant. The fresh shoot weight from seeds of very low phosphate treated mother plant found low compare to all other phosphate and nitrate treatment effects (Fig 21).

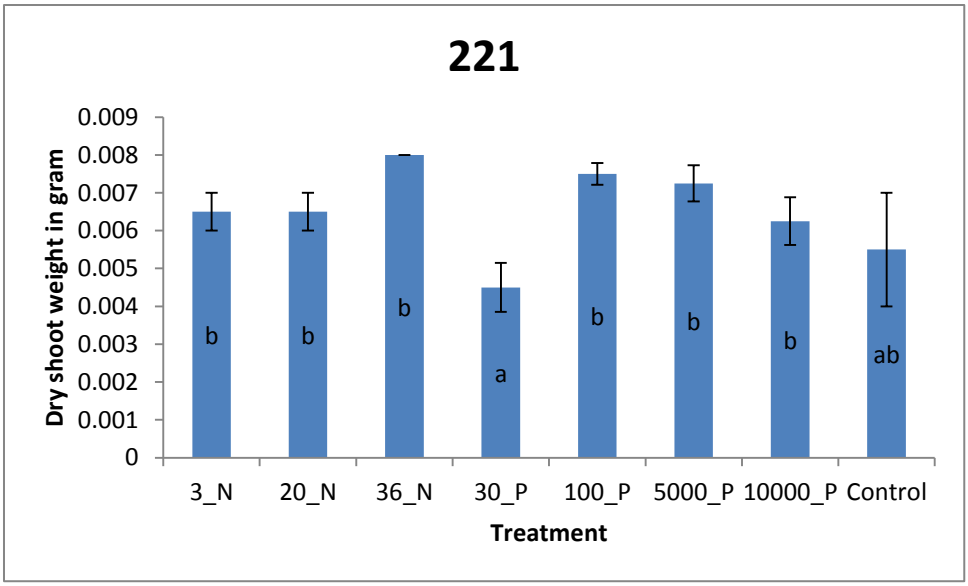

Figure 22 The Means and SE± of dry shoot weight in gram 10 days after growth on copenhagen table in line 221. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05

In line 221 for the dry shoot weight no significant difference found in different treatment except seeds of very low phosphate treated mother plant. The fresh shoot weight from seeds of very low phosphate treated mother plant found low compare to all other phosphate and nitrate treatment effects (Fig 22). In line 221 with respect to fresh and dry root weight no significant difference observed. Data not shown.

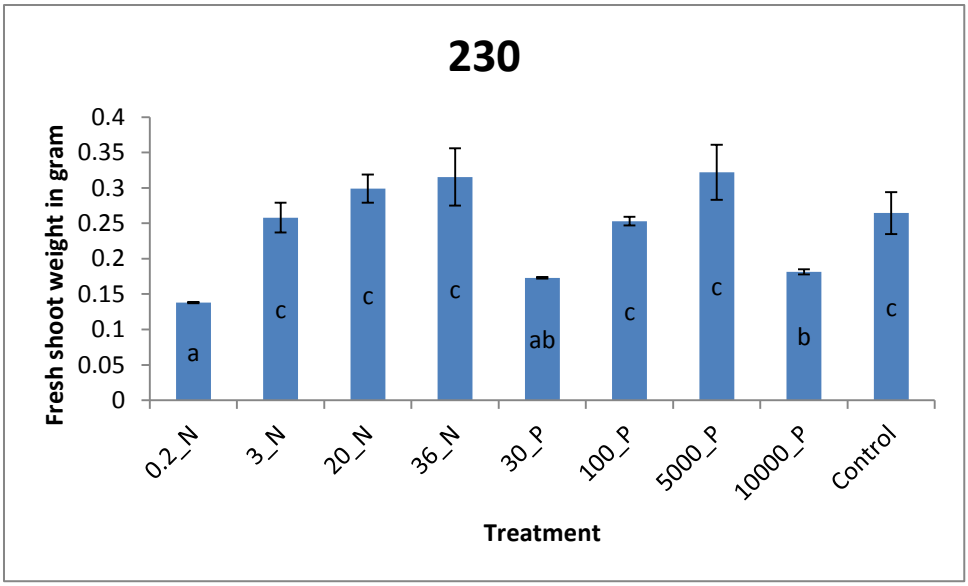

Figure 23 The Means and SE± of fresh shoot weight in gram 10 days after growth on copenhagen table in line 230. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05

In line 230 for the fresh shoot weight, in seeds of very low nitrate treated mother plant less fresh shoot observed compare to seeds of very high phosphate treated mother plant. In seeds of very low nitrate, very low phosphate and very high phosphate treated mother plant less fresh shoot weight founded compare to seeds of low, high and very high nitrate and low and high phosphate treated mother plant and control (Fig. 23).

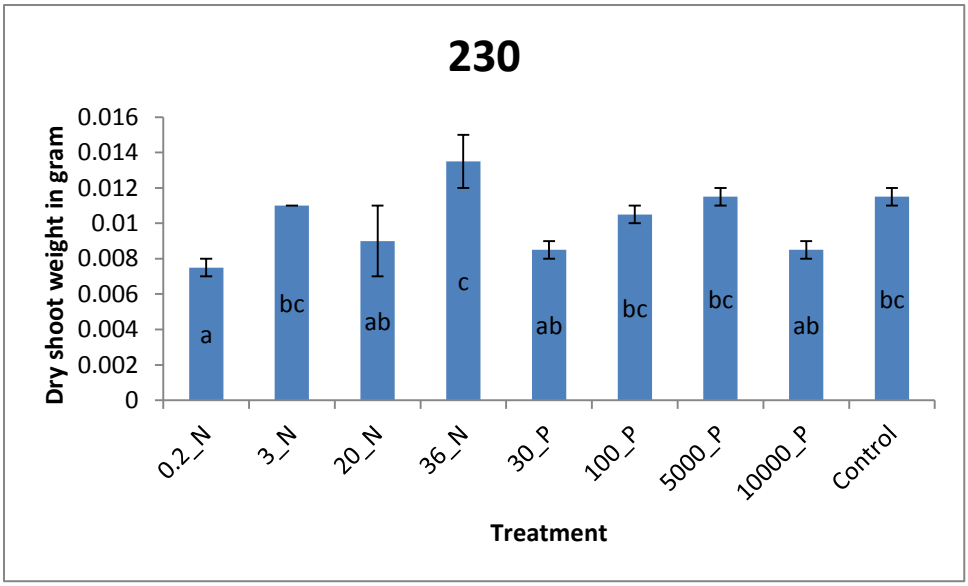

Figure 24 The Means and SE± of dry shoot weight in gram 10 days after growth on copenhagen table in line 230. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05.

In line 230 for the dry shoot weight, low dry shoot weight observed in seeds of very low nitrate treated mother plant compare to seed of low nitrate and high nitrate treated mother plant and low phosphate and high phosphate treated mother plant and control. The dry shoot weight of seeds of very high nitrate treated mother plant higher than seeds of high nitrate and very low phosphate treated mother plant (Fig. 24).

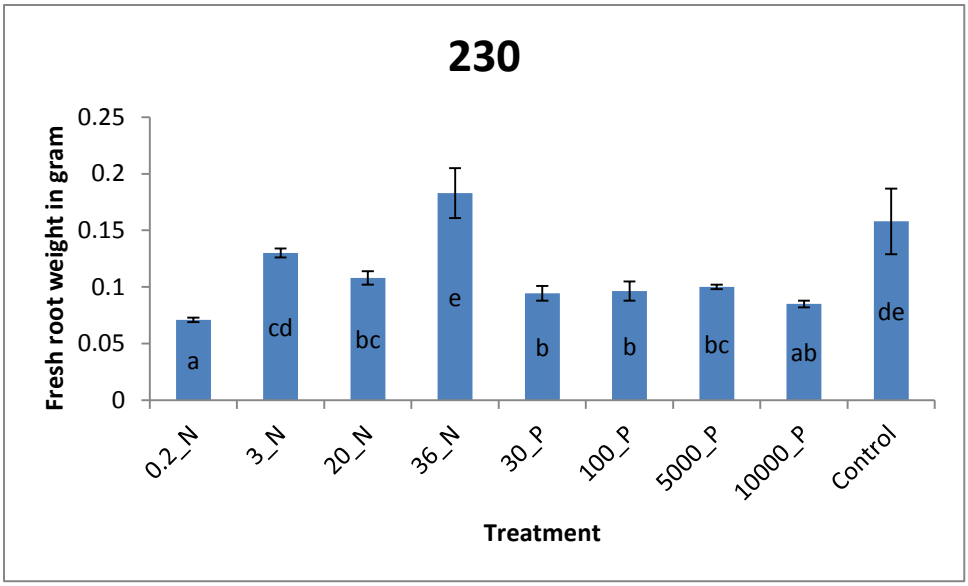

Figure 25 The Means and SE± of fresh root weight in gram 10 days after growth on copenhagen table in line 230. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05.

In line 230 for the fresh root weight in seeds of very high nitrate treated mother plant higher weight observed than seeds of very low nitrate, low nitrate and high nitrate treated mother plant and very low phosphate, low phosphate, high phosphate and very high phosphate treated mother plant. In seeds of control and low nitrate treated mother plant higher weight observed than seeds of very low nitrate and high nitrate treated mother plant and seeds of very low phosphate, low phosphate and very high phosphate treated mother plant. In seeds of very low nitrate treated mother plant low weight observed than seeds of high nitrate, very low phosphate, low phosphate and high phosphate treated mother plant (Fig. 25).

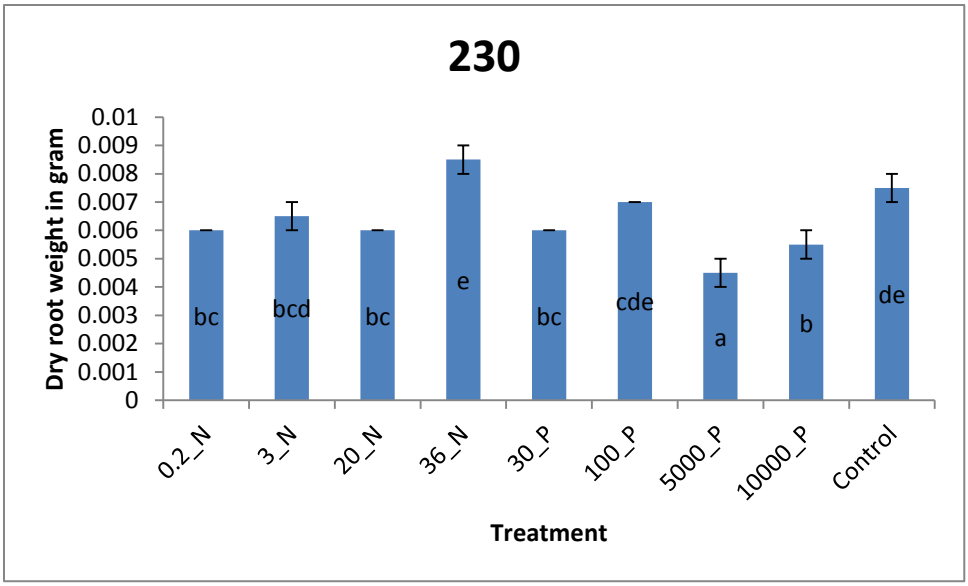

Figure 26 The Means and SE± of dry root weight in gram 10 days after growth on copenhagen table in line 205. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05.

In line 230 for the dry root weight in seeds of very high nitrate treated mother plant higher weight found than very low nitrate, low nitrate and high nitrate treated mother plant and in very low phosphate, high phosphate and very high phosphate treated mother plant. In seeds of low phosphate treated mother plant and control higher weight observed than seeds of high phosphate and very high phosphate treated mother plant. Higher dry root weight observed in seeds of very high phosphate treated mother plant than seeds of high phosphate treated mother plant. No significant difference found in very low nitrate, low nitrate and high nitrate and very low phosphate and very high phosphate treatment (Fig. 26).

In line 254 with respect to fresh and dry shoot weight and fresh and dry root weight no significant effect of different treatment observed. Data not shown.

In line 261 with respect to fresh shoot weight no significant of different treatment observed. Data not shown.

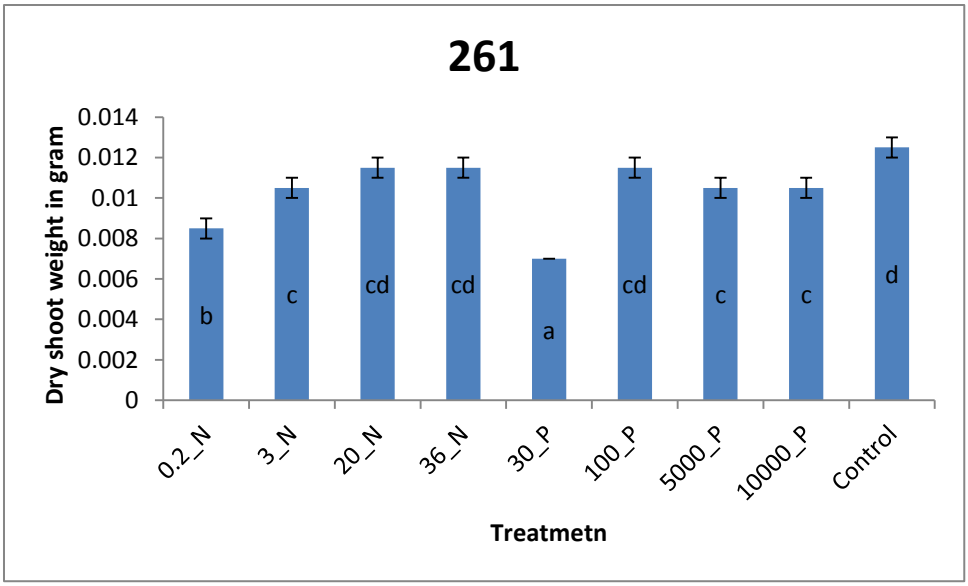

Figure 27 The Means and SE± of dry shoot weight in gram 10 days after growth on copenhagen table in line 261. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05

In line 261 for the dry shoot weight in seeds of control higher dry shoot weight observed than seeds of low and very low nitrate treated mother plant and seeds of very low phosphate, high phosphate and very high phosphate treated mother plant. In seeds of low nitrate, high nitrate and very high nitrate and low phosphate, high phosphate and very high phosphate higher dry shoot weight observed than seeds of very low nitrate and seeds of very low phosphate treated mother plant. In seeds of very low phosphate treated mother plant lower dry shoot weight found than seeds of very low nitrate treated mother plant (Fig 27). In line 261 with respect to fresh root weight no significant of different treatment observed. Data not shown.

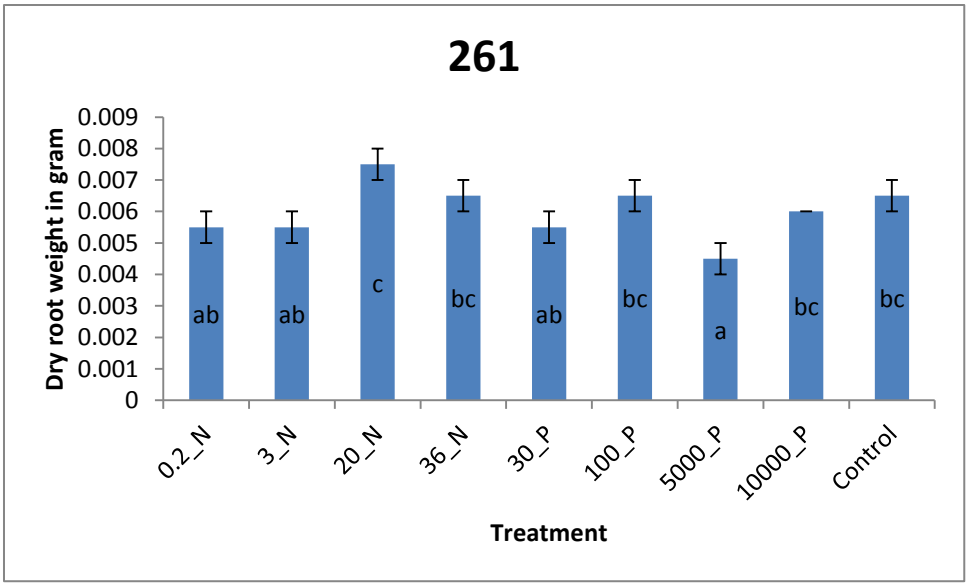

Figure 28 The Means and SE± of dry root weight in gram 10 days after growth on copenhagen table in line 261. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05.

In line 261 for the dry root weight in seeds of high nitrate treated mother plant higher dry root weight observed than seeds of very low nitrate and low nitrate treated mother plant and very low and high phosphate treated mother plant. In seeds of high nitrate, low phosphate and very high phosphate and control higher dry root weight observed than seeds of high phosphate treated mother plant (Fig 28).

In line 290 with respect to fresh shoot weight no significant effect of different treatment observed. Data not shown.

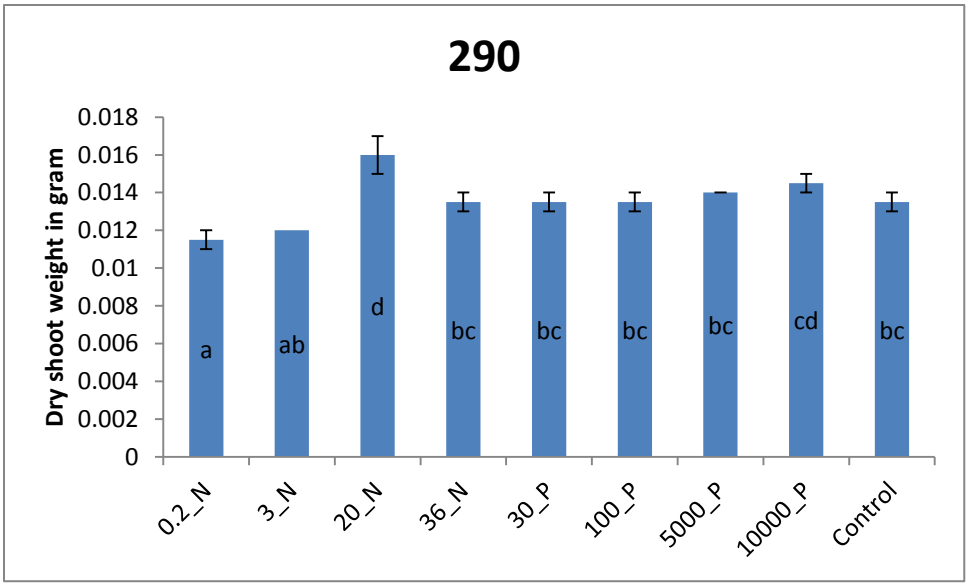

Figure 29 The Means and SE± of dry shoot weight in gram 10 days after growth on copenhagen table in line 290. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05

In line 290 for the dry shoot weight higher dry root weight observed in seeds of high nitrate treated mother plant than very low nitrate, low nitrate and very high nitrate treated mother plant and very low phosphate, low phosphate and high phosphate treated mother plant and control. Lower dry shoot weight observed in seeds very low nitrate treated mother plant than seeds of control, very high nitrate treated mother plant and very low phosphate, low phosphate, high phosphate and very high phosphate treated mother plant (Fig 29). In line 290 with respect to fresh and dry root weight no significant effect of different phosphate and nitrate treatment observed. Data not shown.

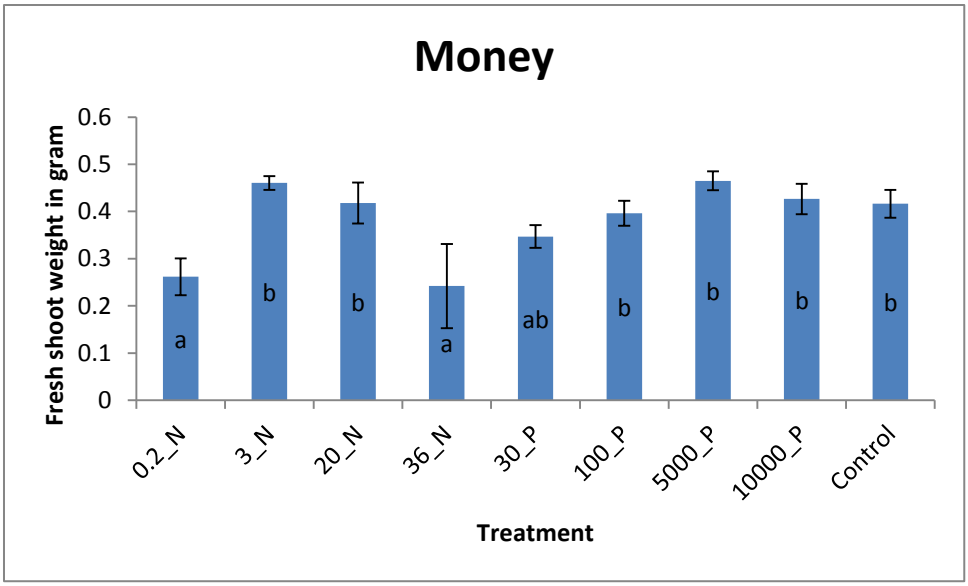

Figure 30 The Means and SE± of fresh shoot weight in gram 10 days after growth on copenhagen table in line money. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05

In line money for the fresh shoot weight lower fresh shoot weight found in seeds of very low nitrate and very high nitrate treated mother plant than seeds of control, low nitrate and high nitrate and low phosphate, high phosphate and very high phosphate treated mother plant (Fig 30).

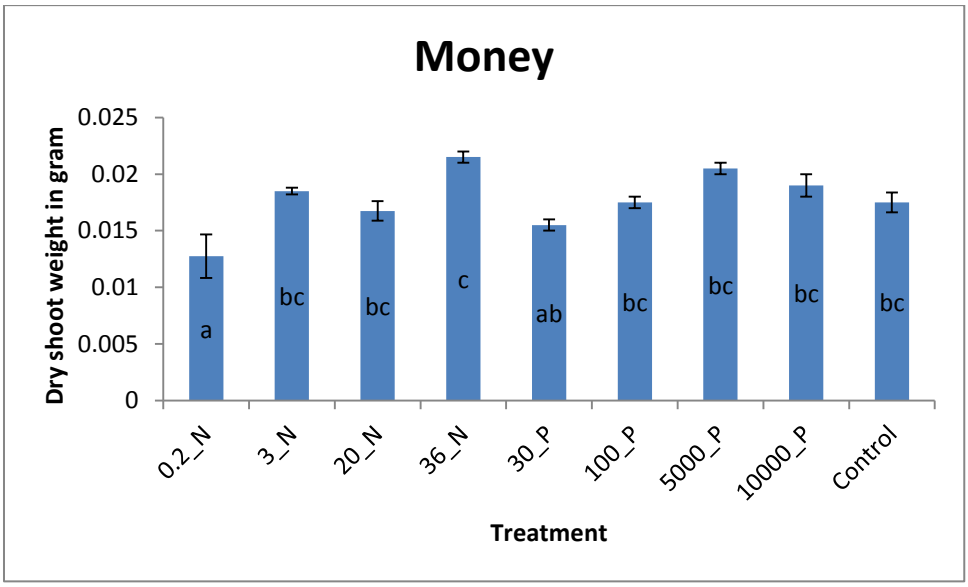

Figure 31 The Means and SE± of dry shoot weight in gram 10 days after growth on copenhagen table in line money. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05

In line money for the dry shoot weight, lowest dry shoot weight observed in seeds of very low nitrate treated mother plant compare other phosphate and nitrate treatment effect. In seeds of very high nitrate treated mother plant dry shoot weight higher than seeds of very low phosphate treated mother plant (Fig 31).

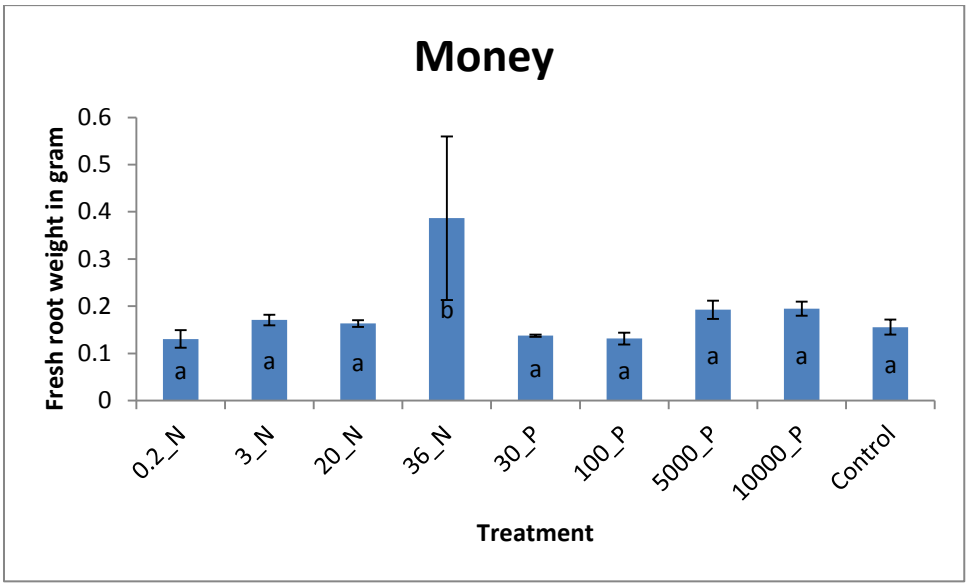

Figure 32 The Means and SE± of fresh root weight in gram 10 days after growth on copenhagen table in line money. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05.

In line money for the fresh root weight only in seeds of very high nitrate treated mother plant higher weight observed than other nitrate and phosphate treatment effect (Fig 32). In line money with respect to dry root weight and no significant effect of different treatment observed. Data not shown.

In line pimp with respect to fresh and dry shoot weight no significant effect of different treatment observed. Data not shown.

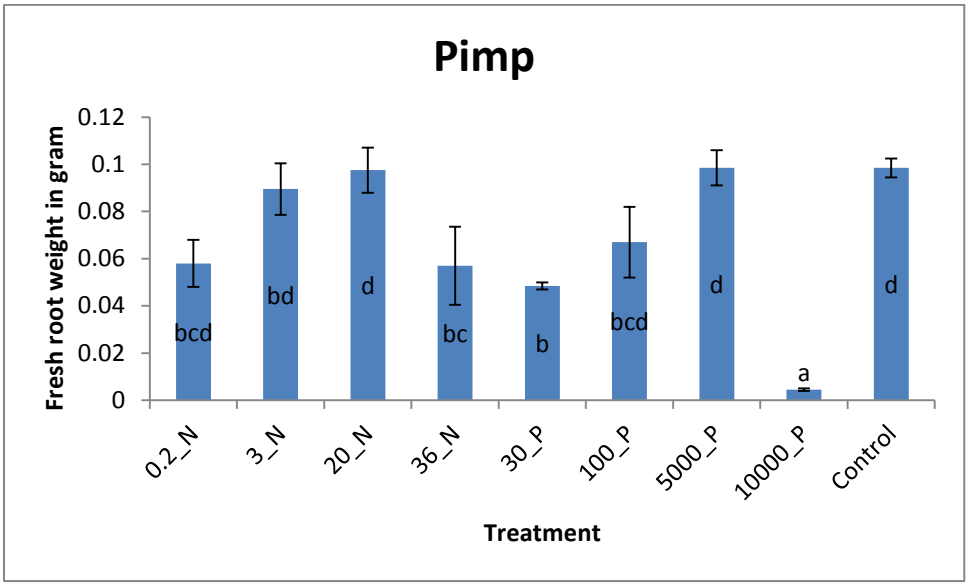

Figure 33 The Means and SE± of fresh root weight in gram 10 days after growth on copenhagen table in line pimp. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05.

In line Pimp for the fresh root weight lowest fresh root weight found in seeds of very high phosphate treated mother plant compare to other phosphate and nitrate treatment. In seed of control, high nitrate and high phosphate treated mother plant higher fresh root weight observed than seed of seed of very high phosphate and very low phosphate treated mother plant (Fig 33). In line pimp with respect to dry root weight and no significant effect of different treatment observed. Data not shown.

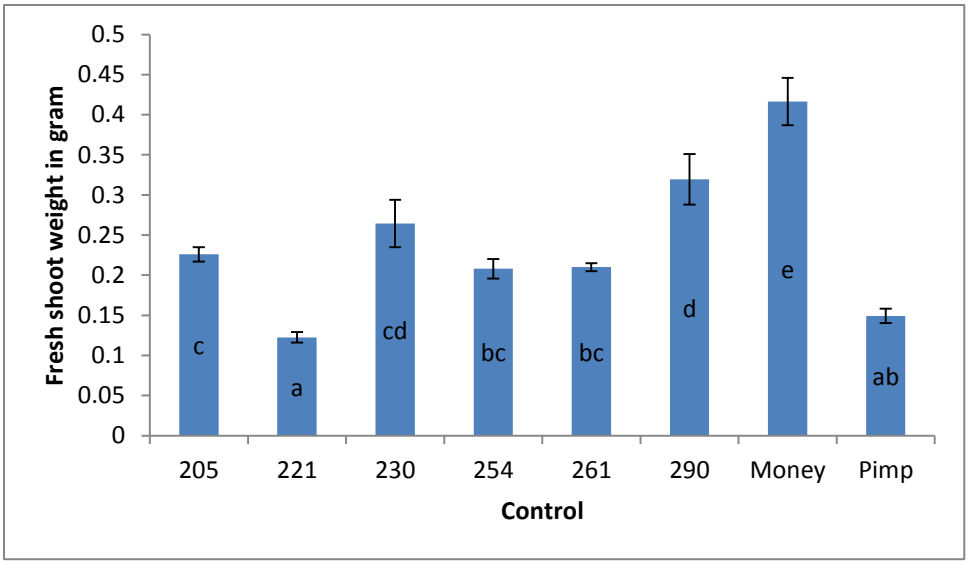

Figure 34 The Means and SE± of fresh shoot weight in gram 10 days after growth on copenhagen table in all genotype under control condition. Different letters indicate statistically significant differences by Fisher's least significant difference (LSD) test; α=0.05.

This is the effect from all line's mother plant grown in control condition. Fresh shoot weight highest in line money and lowest in line 221. This indicates the genotypic variation while seed developed under same control treatment (Fig 34).

	0.2N	3N	20N	30P	100P	5000P	10000P	Control
3N	0.037335							
20N	0.020551	0.973169						
30P	-0.22123	0.81716	0.83426					
100P	0.034715	0.948545	0.945436	0.847626				
5000P	0.070655	0.956794	0.944854	0.753268	0.953768			
10000P	0.254034	0.957841	0.911048	0.744226	0.899156	0.875412		
Control	0.08359	0.991326	0.991591	0.809283	0.951755	0.966524	0.941138	
36N	0.252222	0.532829	0.660153	0.246636	0.513269	0.612569	0.452816	0.627055

Figure 35. Pearson's correlation of different nitrate and phosphate treatments based on fresh shoot weight. Colour represent the degree of correlation, green colour represent very high correlation, yellow represent high correlation, orange represent low or negligible correlation and red represents negative correlation. .

The very low nitrate treatment had minute correlation with treatment high nitrate, low phosphate, high phosphate and control. Weak positive correlation with treatment very high nitrate and very high phosphate and weak negative relationship with very low phosphate. The very low phosphate had high positive correlation with all other treatment except very

high nitrate and very low nitrate. The very high nitrate had a moderate correlation with all the treatments except very low nitrate and very low phosphate treatment. The remaining treatment low nitrate, high nitrate, low phosphate, high phosphate, very high phosphate and control had very strong high positive correlation with each other (Fig. 35).

	0.2N	3N	20N	30P	100P	5000P	10000P	Control
3N	0.712141							
20N	0.847681	0.716657						
30P	0.695393	0.68537	0.797353					
100P	0.717526	0.088052	0.32625	0.518567				
5000P	0.548702	0.783918	0.682371	0.664204	0.121494			
10000P	0.908485	0.729333	0.68108	0.798015	0.426843	0.706186		
Control	0.194039	0.805644	0.35048	0.315478	-0.30063	0.668574	0.447727	
36N	0.7464	0.904865	0.594572	0.720808	0.159678	0.87904	0.882581	0.759699

Figure 36 Pearson's correlation of different nitrate and phosphate treatment based on fresh root weight. Colour represent the degree of correlation, green colour represent very high correlation, yellow represent high correlation, orange represent low or negligible correlation and red represents negative correlation.

The treatment low phosphate had moderate positive correlation with high nitrate and very high phosphate and high correlation with very low nitrate treatment. It had low negative correlation with control and minute positive correlation with high phosphate and very high nitrate. The very low nitrate treatment had strong high positive correlation with high nitrate and very high phosphate, very low with control and high positive correlation with low nitrate, low phosphate, high phosphate and very high phosphate. the low nitrate treatment had very high positive strong correlation with high phosphate, very high phosphate, control and very high nitrate, high positive correlation with high nitrate and very low phosphate and minute positive correlation with low phosphate treatment. The high nitrate had high very high strong positive correlation with very low phosphate and high positive correlation with high phosphate and very high phosphate and low correlation with control and low phosphate treatment. The treatment very low phosphate had high strong correlation with

very high phosphate and very high nitrate, high correlation with low and high phosphate and low correlation with control. The high phosphate had very strong high correlation with very high nitrate and high correlation with very high phosphate and control treatment very high phosphate had moderate correlation with control (Fig. 36).

Discussion

In previous study to know the effect of nitrate and phosphate on seed quality in tomato experiment conducted only using one genotype. In this study we have found the variation in seed quality parameters based on genotypes and treatment.

For germination percentage the effects were not the same in all genotypes. For nitrate treatment in line 261 seed developed under very high nitrate showed less germination. In contrast, line 221, 230 and money showed higher germination in seeds developed under high nitrate conditions. For phosphate treatment we have found higher germination in seeds developed under very low phosphate conditions in line 290 and higher germination in seeds developed under low phosphate. However no significant effect of different phosphate treatments on seed germination was found in line 205, 221, 230, 254, 261 and money. On other hand in previous study by Singh and Kumar on tomato cultivar Arka Abha they did not find significant effect of nitrogen and phosphorous treatment on seed germination [15]. The reason may be they have only used the one cultivar. In our study we have found that higher germination in seed developed under medium phosphate and high nitrate condition. In another study on tomato cultivar Moneymaker showed that the germination rate and seedling emergence was higher in seeds developed under high nitrate and high phosphate conditions [14]. However the experiment was conducted on field condition. For t_{10} and t_{50} we found the contradictory result in line 254 and 290. In line 254 seed developed in very

high phosphate condition require less time to reach, but in 290 seed developed in very low phosphate condition require less time.

For seedling quality also we have found the different effect due to genotype and treatment interaction. For fresh shoot weight in line 221, 254, 261, 290 and pimp no difference in different nitrate treatment was measured. However low fresh shoot weight was measured in line 205, 230 and money from seed developed under very low nitrate condition. In line 254, 261, 290, money and pimp no difference in different phosphate treatment was measured. However in line 205, 221 and 230 low fresh shoot weight measured from seed developed under very low phosphate condition compare to low phosphate, high phosphate and very high phosphate condition. For dry shoot weight In line 205, 221, 254 and pimp no difference in different nitrate treatment was measured. However low dry shoot weight was measured in line 230, 261, 290 and money from seed developed under low nitrate condition compare to low nitrate, high nitrate and very high nitrate condition. In line 230, 254, 290, money and pimp no difference in different phosphate treatment was measured. However In line 205, 221 and 261 lower dry shoot weight was measured from seed developed under low phosphate condition than seed developed under low phosphate, high phosphate and very high phosphate condition.

For fresh root weight in line 221, 254, 261 and 290 no significant effect of different nitrate treatment was measured. However higher fresh root weight observed in line 205, 230, money and pimp from seed developed under very high nitrate condition compare to other very low nitrate, low nitrate and high nitrate condition. In line 221, 230, 254, 261, 290 and money no significant effect of different phosphate treatment was measured. However in line 205 lower fresh root weight measured from seed developed under very low phosphate

condition, but in line pimp lower fresh root weight measured from seed developed under high phosphate and low phosphate condition. For dry root weight in line 205, 221, 254, 290, money and pimp no significant effect of different nitrate treatment was. However in line 230 higher dry root weight observed in seed developed under very high nitrate condition, but in line 261 higher dry root observed in seed developed under high nitrate condition. In line 205, 221, 254, 290, money and pimp no significant effect of different phosphate treatment was measured. However in in line 230 and 261 low dry root weight observed in seed developed under high phosphate condition.

On tomato very few study has been done so far on the effect of seed developed under different nutrient conditions on seed quality. In other crops some studies have shown effect and some not. In an experiment with two verities of *Sorghum bicolor* L., significant effect of seed development under different nitrate and phosphate conditions on seed germination has been found, but only in one variety significant effect on seedling emergence was found. The germination was higher in seeds developed under a combination of normal nitrate and phosphate conditions than without nitrate and phosphate [21]. In a study on six cultivars of rapeseed the author showed the significant effect on germination in seed developed under different nitrogen condition in field experiment. The higher germination in cultivar zafran in high nitrogen condition. However in laboratory conditions significant effect on germination was not found [22]. In study on *Pisum sativum* L., cultivar sprite author conducted two field experiment, in one experiment the higher seed yield was measured from plant developed under high nitrate and phosphate condition but in subsequent experiment no significant effect on seed yield was measured, but in both experiment higher dry weight in seed developed under high nitrate and phosphate treatment observed [23]. However experiment conducted on only one variety. In experiment on three cultivar of cotton (*Gossypium*

hirsutum L.), higher seed cotton yield in plant developed under high nitrogen condition was measured than plant developed in low nitrogen condition [24]. In an experiment on chickpea (*Cicer arientinum* L.) using six cultivar, no significant effect on 1000 grain weight in seed developed under different nitrogen and phosphorous treatment was measured [25]. In field experiment on *Capsicum annuum var. annuum L.* cultivar Anaheim Chili, author not found significant effect of different nitrate treatment on seed quality parameter : germination percentage, seedling weight and field emergence [26]. However the experiment was conducted only on one variety. In field experiment on three variety of okra author did not find any significant effect of nitrogen and phosphorous on progeny seed germination [27]

There is an effect on maternal treatment of different phosphate and nitrate condition. However we have not find in all genotype. Although we found the differences due to different phosphate and nitrate treatment on seed quality, it was genotype dependant. In this study we have only used less replication. In future for further study to reveal more inside we need to increase replication so we can increase statistical power. This knowledge is giving indication that it is possible to identified gens which interact with nitrate and phosphate to improve seed quality.

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