**Experiment-II:**

**Rapid screening of sunflower genotypes for P acquisition- Pot culture study (2012-2016)**

Exp-II a. Protocol to identify the critical P level for solution culture screening of sunflower genotypes (2012-13)

Table 1. Composition of full strength Hoagland's nutrient solution with(+P) and without P (-P) nutrient

|  |  |  |
| --- | --- | --- |
| Different salt solutions of 1*M* strength | Quantity of solution for +P (ml/L) | Quantity of solution for -P (ml/L) |
| KNO3 | 6 | 6 |
| Ca(NO3)2.4H2O | 4 | 4 |
| NH4H2PO4\* | 2 | 0 |
| MgSO4.7H2O | 1 | 1 |
| KCl | 1 | 1 |
| H3BO3 | 1 | 1 |
| MnSO4.H2O | 1 | 1 |
| ZnSO4.7H2O | 1 | 1 |
| H2MoO4 | 1 | 1 |
| Fe-citrate | 1 | 1 |
| NH4Cl  | 0 | 2 |
| Ca(NO3)2.4H2O | 6 | 6 |

\* Volume of 1*M* NH4H2PO4 was adjusted w.r.t. P concentration in the treatments (mgL-1)as 2.0mL required for obtaining full strength P solution(62 mgL-1)

**Results and Discussion**

**Identification of P level critical for initial sunflower growth**

 In Hoagland's nutrient solution, nine levels of phosphorus 0, 2, 3, 4, 5, 6, 7, 8ppm and full strength(62ppm) were maintained to find out the minimum P concentration required to produce significantly lowest dry matter from nearest high level to standardize the critical concentration of P required to develop the protocol for rapid screening of sunflower genotypes in solution culture suitable for identification of changes in root morphology against phosphorus starvation. The data presented in Table 2 indicate the effect of different P levels on growth parameters of sunflower grown in solution culture up to 30 day stage.

Table 2. Effect of P concentrations on growth parameters of sunflower in solution culture at 30 days

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| P levels in nutrient solution(mgL-1) | Shoot length (cm) | Root length (cm) | Shoot weight (g) | Root weight (g) | Total dry matter (g) | Shoot/Root |
| 0 | 16.4 | 32.6 | 0.30 | 0.16 | 0.46 | 1.82 |
| 2 | 24.5 | 31.4 | 1.38 | 0.71 | 2.09 | 1.94 |
| 3 | 25.6 | 32.7 | 1.54 | 0.78 | 2.32 | 1.98 |
| 4 | 26.3 | 31.2 | 1.75 | 0.82 | 2.58 | 2.13 |
| 5 | 29.2 | 26.8 | 2.01 | 0.84 | 2.85 | 2.40 |
| 6 | 28.6 | 28.4 | 2.17 | 0.84 | 3.00 | 2.59 |
| 7 | 28.4 | 32.8 | 2.19 | 0.88 | 3.07 | 2.51 |
| 8 | 29.9 | 29.7 | 2.34 | 0.91 | 3.25 | 2.58 |
| 62(Full strength)  | 31.3 | 30.6 | 2.88 | 1.12 | 4.00 | 2.60 |
| CD (0.05%) | 3.6 | 6.3 | 0.18 | 0.10 | 0.20 | 0.31 |

Shoot length of sunflower seedling was drastically reduced at 4.0 ppm P which was significantly lower than the value obtained at 8.0 ppm, however, it was at par with the growth produced between 3.0 to 7 ppm. The lowest shoot length was noticed due to 2.0 ppm P solution concentration and this level was found to be critical for shoot growth of sunflower and suitable for standardization of protocol for rapid screening of sunflower genotypes. On the other hand, the shoot length due to P levels beyond 4.0 ppm up to full strength had produced similar growth. Similarly, Amit Kumar *et al* (2009) adopted solution culture with sufficient and deficient P levels for screening ground nut lines for identifying P acquisition root traits. It was evident from the results presented in the table 2 where these treatments ( > 2 to < 8 ppm respectively) were statistically at par to each other. It was interesting to note that at lower levels of P, the root length of seedlings was greater compared to sufficient levels (i.e. above 5.0ppm) and this might be due to the response against P starvation in nutrient solution. Further, there was no significant difference in the root length due to P levels. However, the results showed that root dry matter weight was highly influenced by P levels in solution culture. The effect of P levels *viz.,* 0, 2, 3 and 4ppm was on par to each other on shoot/root ratio (Table 2).

 At 30 day growth stage the highest shoot and root dry matter was noticed in full strength solution(62 ppm) while lowest in zero ppm (Fig 2). The minimum growth at 0 ppm may be due to the endosperm nutrient support from the seed. The shoot, root dry weights and total dry matter produced at 2.0ppm P were found to be significantly lowest from the nearest (3.0ppm) concentration of P level in solution. Hence, 2.0 ppm P could be critical level in solution for sunflower for P starvation tolerance. The relationship between P levels in nutrient solution and the total dry matter of sunflower up to 30 days was subjected to best fit and the quadratic equation obtained showed highly significant coefficient of regression value (R2= 0.91) has been depicted in Figure 1. The predicted declined growth of sunflower due to critical P (2.0ppm) was identified between 0 and 3ppm in the graph.

Fig 1. Regression curve for solution P concentration and total dry matter of sunflower

**Phosphorus uptake in sunflower shoot, root and seedling**

 Phosphorus (P) levels in solution culture significantly influenced the shoot, root and plant P content and uptake. The corresponding data has been presented in Table 3. P contents in shoot due to 2.0, 3.0 and 4.0ppm did not differ significantly but were at par to each other however, it was found to be significantly low compared to high levels of P (> 5.0ppm). The treatment with 2.0 ppm recorded lowest P content and corresponding shoot uptake (0.96mgP/pl). Root P content was higher than shoot at 2.0 ppm but corresponding root uptake was lower than shoot which may be ascertained due low root biomass against shoot. The phosphorus uptake by sunflower seedlings at 30 day period was 2.05mgP/plant at 2.0 ppm and 2.47mgP/pl at 3.0 ppm. This could be attributed to lowest total dry matter(2.09g/pl) produced with 2.0ppm P (Table 3). However, the highest P uptake (22.4mgP/pl) was noticed due to full strength nutrient solution. It is imperative that better root-shoot growth and total dry matter might have contributed towards highest uptake.

Table 3. Effect of P levels on P concentration and uptake in sunflower seedlings in solution culture at 30 days

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| P levels in nutrient solution(mg/L) P levels in nutrient solution(ppm) | Shoot P content (%) | Root P content (%) | Shoot uptake(mgP/pl) | Root uptake(mgP/pl) | Plant uptake (mgP/pl) |
| 0 | 0.41 | 0.59 | 0.12 | 0.10 | 0.23 |
| 2 | 0.70 | 1.27 | 0.96 | 0.90 | 2.05 |
| 3 | 0.79 | 1.34 | 1.21 | 1.04 | 2.47 |
| 4 | 0.72 | 1.18 | 1.25 | 0.97 | 2.44 |
| 5 | 0.96 | 1.58 | 1.93 | 1.32 | 3.61 |
| 6 | 1.18 | 1.77 | 2.55 | 1.48 | 4.42 |
| 7 | 1.77 | 1.77 | 3.88 | 1.52 | 5.41 |
| 8 | 1.35 | 1.90 | 3.16 | 1.72 | 5.28 |
| 62(Full strength) | 3.38 | 7.86 | 9.73 | 8.77 | 22.44 |
| CD (0.05%) | 0.13 | 0.51 | 0.35 | 0.64 | 0.84 |

Fig 2. Response of sunflower growth to different P levels in nutrient solution