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In vitro Selection of Drought Stress Tolerance in Rice (*Oryza sativa* L.) var. Jarava

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To develop a drought tolerant variety of Jarava, to a higher level of tolerance by exposing the callus produced from the mature embryo to different levels of PEG (0.0, 0.1, 0.2 and 0.3%) and then selecting the best tolerant cells of the drought and obtaining plants from them was carried out. The plants tested with the same concentrations of PEG, in normal conditions (pots) and evaluated by some indicators of proline, carbohydrate, K, Na and Cl.

The drought affects the soil directly through its effect on the soil composition and indirectly desertification of the land through the impact on the plants. Which leads to the migration of farmers from the rural areas to urban, as a result of the deterioration of their living and health conditions. Affect the destruction of animal wealth, and the surrounding area condition to become hot Xoconostle-Cázares et al. (2011).

The production of one Kg of rice requires 3000- 5000 litres of water, and this amount is three times what needs other plants, which receives 34 - 43% of the world water (Lafitte and Courtois 2002). The mechanism of tolerance of drought in plants is linked to several ways; vary from plant to plant depending on the type of plant. The cell functions under the influence of stress and the ability of cells to withstand the damage resulting from stress. Dynamic organization of events, regulating the vital events of growth under tensile strength helps them to protect the cell itself and get rid of toxicity (Detoxification) resulting from stress (Fang and Xiong 2015, Kumar et al. 2017). The effect of the salinity factor is very similar to the drought factor in the plant. Both are related with the water availability. Increasing salt concentration in the medium reduces available water, Gaxiola et al. (2001). Plant tissue culture technique in laboratory

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can be used to produce line of rice tolerant to drought. This method also produce an operative means carry on advantage of genetic variation in plants, Biswas et al. (2001) and Joshi (2011).

After obtaining the appropriate size of callus, Reddy and Obaid (2017), prepared medium for drought to the above mentioned medium was modified with PEG (1, 2 and 3%). The medium was distributed to 200 ml/bottle, followed by the sterilization process, and then left for three days to ensure sterilization efficiency, 50 mg of callus was transferred to each bottle and incubated in the same conditions (25 ± 2^{0}C in dark). After the growth of callus (three weeks), soft weight of callus was measured. The callus was weighed with the bottles in the different PEG concentrations, 10 replicates are used for experiment, with a tube placed with medium only to control the weight loss resulting from evaporation, after two weeks they were weighed with containers and callus salts and repeated this process for 8 weeks. After 8 weeks of growth, the callus samples from each treatment were taken and dried in an electric oven at 65^{0}C for 48 hrs to be prepared for testing and measuring ionic content, carbohydrates and proteins. After acclimatization of plantlet to the field conditions, they were tested to drought tolerance by leaving to drought tolerance by leaving them for two weeks without irrigation. During this period, the plantlets were evaluated for morphological and chemical indicators compared with control, with normal irrigation.

Table 1. Effect of the different concentrations **of PEG on callus and plants** development**.**

PEG	Evaluation of indicators of drought (Mean ± SE)				
conc. (%)	Proline	Carbohydrate	K	Na	Cl
0.0	1.08 ± 0.04	4.93 ± 0.04	6.64 ± 0.09	0.33 ± 0.12	2.34 ± 0.27
1.0	1.37 ± 0.06	6.56 ± 0.04	7.22 ± 0.87	0.36 ± 0.06	1.98 ± 0.12
2.0	1.42 ± 0.02	6.85 ± 0.09	7.68 ± 0.12	0.38 ± 0.02	1.22 ± 0.11
3.0	1.55 ± 0.05	7.45 ± 0.12	8.21 ± 0.76	0.41 ± 0.11	0.96 ± 0.43
PEG	Evaluation of indicators of drought (Mean ± SE)				
conc. (%)	Proline	Carbohydrate	K	Na	Cl
0.0	2.43 ± 0.54	1.08 ± 0.30	9.7 ± 0.9	2.33 ± 0.31	2.31 ± 0.03
1.0	4.22 ± 0.43	2.37 ± 0.37	9.4 ± 0.34	1.76 ± 0.33	1.54 ± 0.19
2.0	6.51 ± 0.26	4.42 ± 0.16	8.6 ± 0.23	0.87 ± 0.20	1.01 ± 0.31
3.0	7.3 ± 0.45	6.55 ± 0.23	8.2 ± 0.12	0.45 ± 0.31	0.78 ± 0.09

In (Table 1 and Fig. 1), the effect of several concentrations of PEG on callus after 8 weeks of inoculation it on the MS, media we noted increased the concentration of proline with increasing the amount of PEG% 1.08 mg/g in the control to 1.42 mg/g, in the treatment 2% of PEG. The concentration of carbohydrates increased from 4.93 mg/g in the control, to 6.85 mg/g in the concentration 2% PEG. Potassium (K) played an important role in drought tolerance where the concentration of PEG% increased, from

6.64 µg/50 mg of potassium in the control and treatment it is 8.2 µg/50 mg in the 2% PEG concentration.

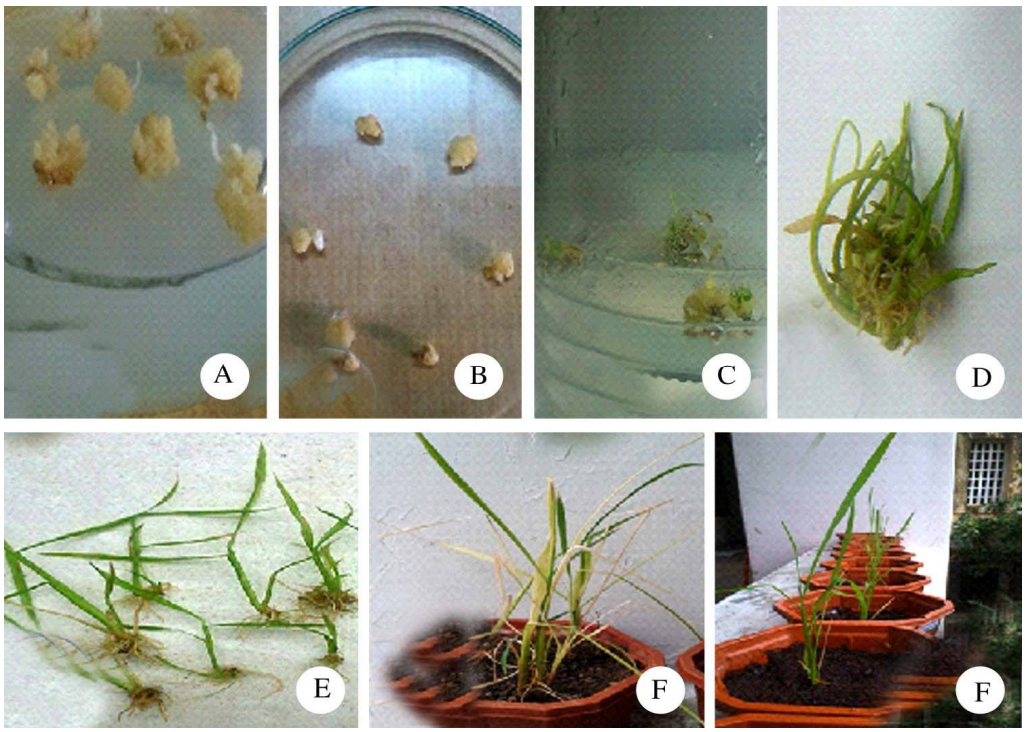

Fig. 1. Regeneration of plants from drought tolerant callus and plants under drought condition. A. Induction callus from matured embryo, B. Callus for drought tolerant, C. Drought tolerant callus convert to green callus and start view plantlet, D. Development of plantlets from callus, E. Whole plant ready to hardening stage, F. Normal plants and G. Drought tolerant plants.

But the accumulation of (Na) was not significantly increased, where it ranged between 0.33 µg/50 mg in the control to 0.38 µg/50 mg in 2% of PEG concentration. While the concentration of chlorine decreased from 2.34 µg/50 mg in the control and in treatment to 1.22 µg/50 mg in the concentration 2% of PEG.

Noted from (Table 1 and Fig.) that the drought conditions affected in the plants. It is noted that the indicators of plant tolerance for drought by increasing the content of proline in the plant tissues, when increasing the concentration of PEG%, from 2.42 mg/g in the control, in treatment it is 6.51 mg/g in concentration PEG 2%, carbohydrates also played an effective role in drought tolerant plants, their concentration increased from 1.08 mg/g in the control in treatment it is 4.42 mg/g in 2% of PEG.

As for the effect of dehydration on the elements K, Na and Cl the concentration of all studied elements decreased from control treatment (9.7, 2.33 and 2.31) µg per 50 mg respectively, to (8.6, 0.87 and 1.01) µg/50 mg, respectively.

Hargurdeep and Sain (1996) reported the effect of drought on rice under controlled conditions, while reducing the proportion of water to 20%, has been obtained from the plant and got the tolerance of stuttering during sugar to starch and also found between the demolition of sugar and starch accumulation.

The proline content will be increased with water stress and the synthesis of proline from the glutamine, proline is indicator for drought stress, has been shown by study Handa et al. (1986). When exposing the cells of the plant to the conditions of drought using PEG the cells gradually swell because of their response and the proline will be accumulated quickly in the cells, because the role of proline in protecting cells from dehydration.

The ions are affected by increasing the concentration of salt stress in a striking manner. Increasing the ions Na^+ and Cl^- by increasing the PEG concentration to the medium. In most of the plants observed increasing the concentration of potassium K^+ with increased drought condition, Kumar and Khare (2016). That they studied the effect of ions, on each ion separately, detected their effect on the wet and dry weight. But there was no significant difference between ions Na^+ and Cl^-. The ions tress caused by plants biomass is a well documented phenomenon, Khare et al. (2014), Ranjit et al. (2015).

In the present investigation, the callus screened under drought condition with PEG, and developed total plants, the proline content estimated in tissue level of callus and plants, the amount of proline is increased, than the control is significant manner, proline one of the amino acid highly soluble and it changes osmotic concentration in cytoplasm of the plants. And also carbohydrates increased. The potassium and sodium concentration increased in callus but not in chlorine ion. The sodium, potassium and chlorine ions decreased in plants. The selected plants can be released as new varieties.

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