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SEED GERMINATION AND EARLY GROWTH OF PHYSIC NUT SEEDLINGS UNDER SALINITY STRESS

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ABSTRACT - Soil salinity is one of the major abiotic stresses that limit seed germination and plant growth, causing yield loss in crop species. The phytotoxic effects of salt stress on seed germination and early growth of physic nut (*Jatropha curcas* L.) seedlings were investigated in the present study. Physic nut seeds were subjected to five levels of salinity [0 (control); 25; 50; 75 and 100 mmol L^{-1} of NaCl], using the solution-paper method, with five replications. The experimental unit consisted in a germitest paper roll with 25 physic nut seeds. Salt treatments were evaluated based on the following tests: first count of germination test (7 days), total germination (12 days) and early growth of physic nut seedlings (12 days). Results showed that the addition of 100 mmol L^{-1} of NaCl reduced the first count of germination test (45%), total germination (50%), shoot length (42%), root length (59%), shoot dry matter (34%) and root dry matter (44%) of physic nut seedlings compared to the NaCl-free control. The exposure to high salt concentrations severely restricted the seed germination and early growth of physic nut seedlings. These data suggest that physic nut crop is a moderately sensitive species to high salinity levels during the stage of seed germination and early seedling growth.
Key words: *Jatropha curcas*, osmotic stress, salt stress.

GERMINAÇÃO E CRESCIMENTO INICIAL DAS PLÂNTULAS DE PINHÃO-MANSO EM CONDIÇÕES DE SALINIDADE

RESUMO - A salinidade é um dos principais estresses abióticos que limitam a germinação das sementes e o desenvolvimento das culturas em solos agrícolas. Este estudo teve como objetivo avaliar os efeitos do estresse salino na germinação das sementes e no crescimento inicial das plântulas de pinhão-manso (*Jatropha curcas* L.). As sementes de pinhão manso foram submetidas a cinco níveis de salinidade [0 (controle), 25, 50, 75 e 100 mmol L^{-1} de NaCl], com cinco repetições. As unidades experimentais foram constituídas de rolos de papel germitest com 25 sementes de pinhão manso. Os tratamentos foram avaliados com base nos seguintes testes: primeira contagem da germinação (7 dias), germinação (12 dias) e crescimento inicial das plântulas de pinhão manso (12 dias). A adição de 100 mmol L^{-1} de NaCl reduziu a primeira contagem da germinação (45%), a germinação final (50%), comprimento da parte aérea (42%), comprimento das raízes (59%), matéria seca da parte aérea (34%) e a matéria seca das raízes (44%) de pinhão manso em comparação as plântulas sem exposição ao estresse salino. A exposição das sementes a altos níveis de salinidade reduziu severamente a germinação das sementes e o crescimento inicial das plântulas de pinhão manso. Estes dados sugerem que o pinhão-manso é uma espécie moderadamente sensível à alta concentração de NaCl na solução durante a fase de germinação das sementes e crescimento inicial.
Palavras-chave: *Jatropha curcas*, estresse osmótico, estresse salino.

INTRODUCTION

Salinity caused by excessive salts in the soil solution or in irrigation water is one of the abiotic stresses that most limit plant growth and development in arid, semiarid and other areas of the world (ZHU, 2001). Currently, the world population growth and economic development have resulted in a growing expansion of degraded soil areas by salinity as consequence of the use of marginal lands and inadequate irrigation management. Twenty percent of the world's cultivated land, 7% of the total world land and 50% of the irrigated land are affected by high salt content (ZHU, 2001).

A soil is considered saline when the electrical conductivity of the extract from the water-saturated soil (ECe) is ≥ 4 dS m^{-1} (RICHARDS, 1954). An ECe of 4 dS m^{-1} is equivalent to about 40 mmol L^{-1} of NaCl. However, growth of many plant species is affected by soil with an ECe less than 4 dS m^{-1}. Thus, Rogers et al. (2005) grouped the soil salinity into three categories, i.e. high salinity (ECe > 8 dS m^{-1}), moderate salinity (ECe between 4 and 8 dS m^{-1}) and low salinity (ECe between 2 and 4 dS m^{-1}).

Adverse effects of salt stress on plant growth are due to ionic imbalance, osmotic stress, specific ion toxicity, oxidative stress (MUNNS, 2002) and molecular

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disorders (MUNNS; TESTER, 2008). Salinity reduces cell turgor and depresses rates of root and leaf elongation (FRICKE et al., 2006), suggesting that environmental salinity acts primarily on water uptake. Excessive salt concentrations in the soil solution also changes the ability of plants to absorb, transport and utilize the nutrients needed for their growth and development (FEIJÃO et al., 2011; PARIDA; DAS, 2005). Nutritional imbalance caused by salinity is mainly due to the reduction of uptake and assimilation of essential nutrients by the plant such as nitrogen (N), potassium (K) and calcium (Ca) (ZHU, 2001).

Salinity affects plant growth at all developmental stages; however, sensitivity varies from one growth stage to another. Seed germination is a critical stage of the plant life cycle and improved tolerance to high salinity levels could improve the stability of crop yield in saline soils. However, the effects of salinity stress on germination and establishment of physic nut (*Jatropha curcas* L.) seedlings are incipient and inconclusive (ANDRÉO-SOUZA et al., 2010).

Physic nut is an important undomesticated perennial plant that has received special attention in recent years due to its high seed oil concentration with excellent quality for utilization in biodiesel production (KUMAR; SHARMA, 2008). This species is widely distributed in the arid and semiarid areas of South America and in all tropical regions (KING et al., 2009), and therefore subject to grow in salinity conditions. The objective of this study was to evaluate the effects of salt stress on seed germination and early growth of physic nut (*Jatropha curcas* L.) seedlings.

MATERIAL AND METHODS

The experiment was conducted at Ourinhos, São Paulo, Brazil (24º 55' 20" S, 49º 54' 24" W, and altitude of 480 m). Physic nut seeds, collected directly from the treetop of a plant population in Marechal Cândido Rondon, Paraná, Brazil (24º 31’ S, 54º 01’ W), were previously selected taking into account the seed size and weight. Afterwards, seeds were surface sterilized for 5 minutes with sodium hypochlorite solution (2%, v/v) and subjected to five levels of salinity [0 (control); 25; 50; 75 and 100 mmol L^{-1} of NaCl], using the solution-paper method.

The experimental design was completely randomized with five replications. The experimental unit consisted in a germitest paper roll with 25 seeds of physic nut.

Germination test: 25 seeds were distributed in two sheets of germitest paper, properly moistened with the salt solution of each treatment, in a volume equivalent to 2.7 times the weight of the dry paper. The seeds then were covered with a third sheet of paper and rolled up. The five rolls, corresponding to the replicates, were grouped by treatment and placed in the germinator at 25 ºC in an upright position to germinate. Evaluations were performed at 7 (first count of germination test) and 12 days (total germination percentage) after the test installation as previously described by Oliveira et al. (2014). The results expressed in percentage of normal seedlings, according to the recommendations of Seed Analysis Rules (BRASIL, 2009).

Shoot and root length: the shoot and root length was measured in 10 normal seedlings randomly obtained after count of the total germination (14 days) using meter scale. The results were expressed in centimeter (cm).

Shoot and root dry matter: 10 normal seedlings randomly obtained at 14 days were separated into shoot and roots. The plant parts were removed carefully, dried in a forced air circulation oven for four days at 65 ºC, and then weighed. The results were expressed in mg per seedling.

The normality of data was previously tested by the Kolmogorov-Smirnov test and then data were submitted to analysis of polynomial regression, and significant equations ($p \leq 0.05$) with the higher coefficient of determination were adjusted. All analyses were performed using SigmaPlot 11.0 software for Windows (Systat Software, Inc., San Jose, CA, USA).

RESULTS AND DISCUSSION

The first count of germination test (7 days) and total germination percentage (12 days) of physic nut seeds decreased progressively with increasing NaCl levels (Figure 1). First count of germination test was reduced from 57.3% in the control treatment (NaCl-free) to a minimum of 31.3% when the seeds were exposed to 100 mmol L^{-1} of NaCl, indicating mean reduction of 45% (Figure 1A).

Total germination percentage was reduced from 81.3% in the NaCl-free control to a minimum of 40.3% when the seeds were exposed to 100 mmol L^{-1} of NaCl, indicating mean reduction of 50% (Figure 1B). When the seeds were exposed to high salinity levels, germination was drastically reduced. The data suggest that physic nut is a moderately sensitive species to the negative effects of high salt levels during the phase of seed germination, confirming the results reported by Andréo-Souza et al. (2010).

Seed germination is a critical stage of the plant cycle and improved tolerance of high salt levels could improve the stability of crop yield under salinity conditions. Andréo-Souza et al. (2010) showed that physic nut seeds have their delayed germination process when subjected to salt stress in the imbibition stage. According to Rafiq et al. (2006), the salt concentration can completely inhibit seed germination at higher levels or induces a state of dormancy at low levels, as well as reduce imbibition of water because of lowered osmotic potentials of the medium and causes changes in metabolic activity. Salt stress affects seed germination through osmotic effects, ion toxicity or a combination of the two effects (BAJJI et al., 2002; SOSA et al., 2005; ZHANG et al., 2010).

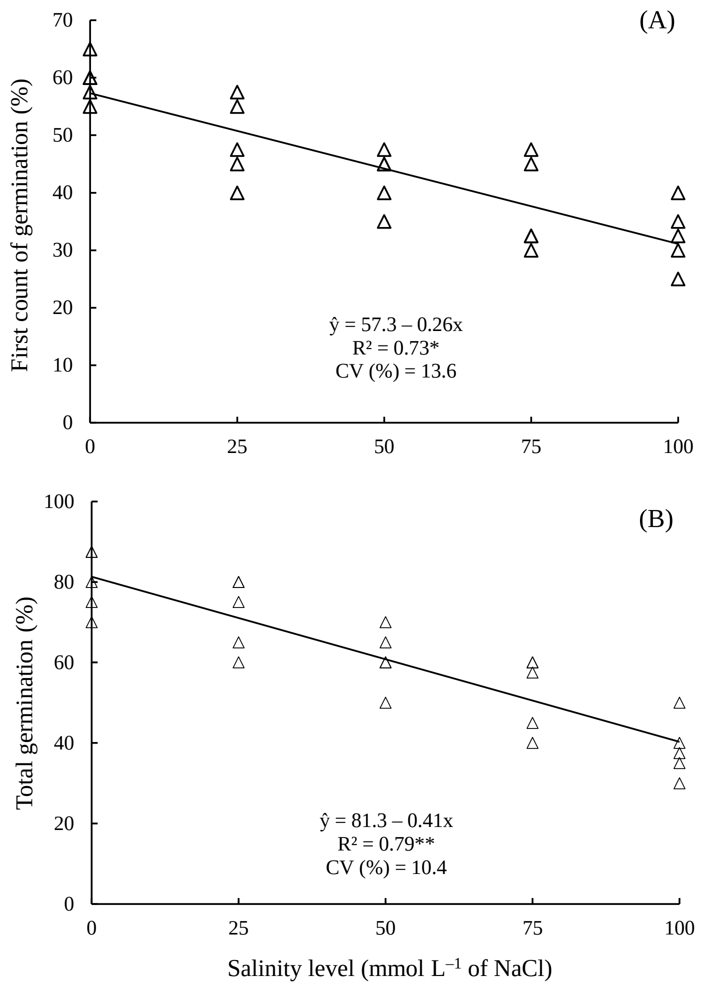

FIGURE 1 - Effects of salt stress on the first count of germination test – 7 days (A) and total germination percentage – 12 days (B) of physic nut (*Jatropha curcas* L.) seeds. * and **: statistical significance at 5% and 1%, respectively.

The growth of physic nut seedlings was negatively affected by the NaCl levels (Figure 2). Shoot length reduced from 19.2 cm in the NaCl-free control to minimum of 11.2 cm with the addition of 100 mmol L^{-1} of NaCl, indicating mean reduction of 42% (Figure 2A). Shoot dry matter reduced from 106.2 mg per seedling in the NaCl-free control to minimum of 70.5 mg per seedling when the seedlings were exposed to 100 mmol L^{-1} of NaCl, indicating mean reduction of 34% (Figure 2C). According to Munns and Tester (2008), excessive salt concentrations in the soil reduce the solution water potential, causing toxic effects and injuries and disorders in the metabolism of plants. Pereira and Lopes (2011) found that the reduction of osmotic potential drastically reduced physic nut seed germination and seedling establishment. Those authors observed that the osmotic potential of -0.2 MPa resulted in the lowest germination index and early seedling growth.

Root length and root dry matter of physic nut seedlings decreased progressively with increasing of NaCl levels. Root length was reduced from 11.4 cm in the NaCl-free control to a minimum of 4.7 cm with the addition of 100 mmol L^{-1} of NaCl, indicating a reduction of 59% (Figure 2B). Root dry matter was reduced from 20.3 mg per seedling in the NaCl-free control to a minimum of 11.3 mg $seedling^{-1}$ in the concentration of 100 mmol L^{-1} NaCl, indicating a reduction of 44% (Figure 2D). Jamil et al. (2006) found that increased salinity caused a significant reduction in germination percentage, germination rate, and early seedling growth. Andréo-Souza et al. (2010) concluded that there is reduction in the physic nut growth when the seedlings are subjected to NaCl solution with electrical conductivity (ECe) ≥ 6 dS m^{-1} (i.e., equivalent to about 60 mmol L^{-1} of NaCl). However, to obtain a proper crop yield under salinity stress, insuring physic nut seed germination and suitable seedling establishment are necessary.

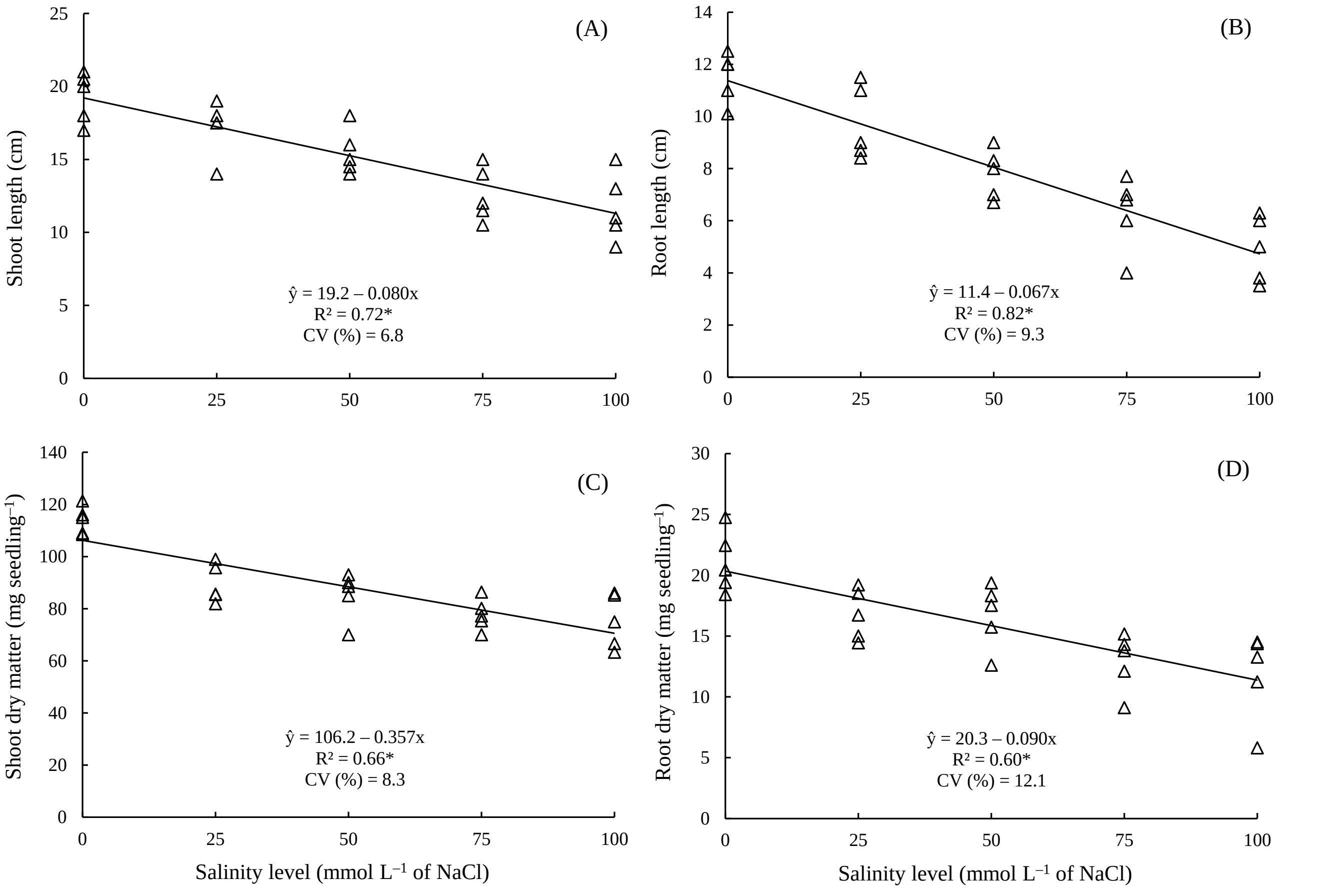

FIGURE 2 - Effects of salt stress on the shoot length (A), root length (B), shoot dry matter (C) and root dry matter (D) of physic nut (*Jatropha curcas* L.) seedlings. *: statistical significance at 5%.

CONCLUSIONS

Salt stress decreased seed germination and length of physic nut seedlings at early growth stage.

The physic nut is a moderately sensitive species to high salt concentrations during the stage of seed germination and early seedling growth.

REFERENCES

ANDRÉO-SOUZA, Y.; PEREIRA, A.L.; SILVA, F.F.S.; RIEBEIRO-REIS, R.C.; EVANGELISTA, M.R.V.; CASTRO, R.D.; DANTAS, B.F. Efeito da salinidade na germinação de sementes e no crescimento inicial de mudas de pinhão-manso. **Revista Brasileira de Sementes**, v.32, n.2 p. 83-92, 2010.

BAJJI, M.; KINET, J.M.; LUTTS, S. Osmotic and ionic effects of NaCl on germination, early seedling growth, and ion content of *Atriplex halimus* (Chenopodiaceae). **Canadian Journal of Botany**, v.80, n.4, p.297-304, 2002.

BRASIL. Ministério da Agricultura, Pecuária e Abastecimento. **Regras para análise de sementes**. Brasília: Mapa/ACS, 2009. 395p.

FEIJÃO, A.R.; SILVA, J.C.B.; MARQUES, E.C.; PRISCO, J.T.; GOMES-FILHO, E. Efeito da nutrição de nitrato na tolerância de plantas de sorgo sudão à salinidade. **Revista Ciência Agronômica**, v.42, n.3, p.675-683, 2011.

FRICKE, W.; AKHIYAROVA, G.; WEI, W.X.; ALEXANDERSSON, E.; MILLER, A.; KJELLBON, P.O.; RICHARDSON, A.; WOJCIECHOWSKI, T.; SCHEIBER, L.; VESELOV, D.; KUDOYAROVA, G.; VOLKOV, V. The short-term growth response to salt of the developing barley leaf. **Journal of Experimental Botany**, v.57, n.6, p.1079-1095, 2006.

JAMIL, M.; LEE, D.B.; JUNG, K.Y.; ASHRAF, M.; LEE, S.C.; RHA, S.E. Effect of salt (NaCl) stress on germination and early seedling growth of four vegetable species. **Journal of Central European Agriculture**, v.7, n.2, p.273-282, 2006.

KING, A.J.; HE, W.; CUEVAS, J.A.; FREUDENBERGER, M.; RAMIARAMANANA, D.; GRAHAM, I.A. Potential of *Jatropha curcas* as a source of renewable oil and animal feed. **Journal of Experimental Botany**, v.60, n.10, p.2897-2905, 2009.

KUMAR, A.; SHARMA, S. An evaluation of multipurpose oil seed crop for industrial uses (*Jatropha curcas* L.): a review. **Industrial Crops and Products**, v.28, n.1, p.1-10, 2008.

MUNNS, R. Comparative physiology of salt and water stress. **Plant, Cell and Environment**, v.25, n.2, p.239-250, 2002.

MUNNS, R.; TESTER, M. Mechanisms of salinity tolerance. **Annual Review of Plant Biology**, v.59, n.7, p.651-681, 2008.

OLIVEIRA, G.L.; DIAS, D.C.F.S.; HILST, P.C.; SILVA, L.J.; DIAS, L.A.S. Standard germination test in physic nut (*Jatropha curcas* L.) seeds. **Journal of Seed Science**, v.36, n.3, p.336-343, 2014.

PARIDA, A.K.; DAS, A.B. Salt tolerance and salinity effects on plants: a review. **Ecotoxicology and Environment Safety**, v.60, n.6, p.324-349, 2005.

PEREIRA, M.D.; LOPES, J.C. Germinação e desenvolvimento de plântulas de pinhão manso sob condições de estresse hídrico simulado. **Semina: Ciências Agrárias**, v.32, supl.1, p.1837-1842, 2011.

RAFIQ, S.; IQBAL, T.; HAMEED, A.; ZULFIQAR-ALI, R.; RAFIQ, N. Morphobiochemical analysis of salinity stress response of wheat. **Pakistan Journal of Botany**, v.38, n.5, p.1759-1767, 2006.

RICHARDS, L.A. **Diagnosis and improvement of saline and alkali soils**. Washington DC: United States Department of Agriculture, 1954. 160p. (Handbook no. 60).

ROGERS, M.E.; CRAIG, A.D.; MUNNS, R.E.; COLMER, T.D.; NICHOLS, P.G.H.; MALCOLM, C.V.; BARRETT-LENNARD, E.G.; BROWN, A.J.; SEMPLE, W.S.; EVANS, P.M.; COWLEY, K.; HUGHES, S.J.; SNOWBALL, R.; BENNETT, S.J.; DEAR, B.S.; EWING, M.A. The potential for developing fodder plants for the salt-affected areas of southern and eastern Australia. An overview. **Australian Journal of Experimental Agriculture**, v.45, n.4, p.301-329, 2005.

SOSA, L.; LLANES, A.; REINOSO, H.; REGINATO, M.; LUNA, V. Osmotic and specific ion effects on the germination of *Prosopis strombulifera*. **Annals of Botany**, v.96, n.3, p.261-267, 2005.

ZHANG, H.; IRVING, L.J.; MCGILL, C.; MATTHEW, C.; ZHOU, D.; KEMP, P. The effects of salinity and osmotic stress on barley germination rate: sodium as an osmotic regulator. **Annals of Botany**, v.106, n.6, p.1027-1035, 2010.

ZHU, J.K. Plant salt tolerance. **Trends Plant Science**, v.6, n.2, p.66-71, 2001.