

RESPONSE OF SOME CANOLA VARIETIES TO SALINITY TOLERANCE USING TISSUE CULTURE TECHNIQUE

By

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ABSTRACT

Nahid Abdelaty Ali Morsi: Response of Some Canola Varieties to Salt Tolerance Using Tissue Culture Technique. Unpublished Ph.D. Thesis, Department of Agronomy, Faculty of Agriculture, Ain Shams University, 2015.

This work was carried out during 2011- 2015 in the laboratories of Cell Research Department (CRD), Field Crops Research Institute (FCRI), Agricultural Research Center (ARC), Giza, as well as, in the greenhouse of Agronomy Department, Faculty of Agriculture, Ain Shams University, Egypt, to study the variation among canola varieties in response to salt tolerance through tissue culture propagation. This work was aimed to establish a regeneration protocol for canola varieties, production of salt-tolerance plants through selection of salt-tolerance callus *via* somaclonal variation and subsequent regeneration of plantlets from such callus, identifying the genetic variations among the used varieties and the newly developed salt tolerant genotypes produced *via* somaclonal variation using RAPD and ISSR banding patterns.

Through evaluation of 22 varieties of canola, Bingo and Torpe were selected as highly salt tolerant varieties while, Conny and Siberian as susceptible varieties. Those four varieties were used for establishment a regeneration protocol. The results indicated that MS medium supplemented with 2 mg/l 2,4-D was the successive medium for callus formation and shoot regeneration was obtained from MS medium supplemented with 5 mg/l BA + 0.05 NAA, while, 1mg/l IBA was the better growth regulator for rooting.

For *in vitro* selection of salt tolerant calli, Torpe variety overcome the others in calli survival percentages under 8000, 12000 and 16000 ppm NaCl concentrations and callus of Siberian variety hold out under this concentration despite that it was within the susceptible group for salinity. The results of shoot frequency from tolerant calli cleared that Torpe variety recorded the highest value of shoot percentage followed by Bingo variety, whereas, Conny followed by Siberian varieties were recorded the lowest values. At 16000 ppm NaCl concentration only tolerant calli of Torpe variety was able to initiate shoot under this concentration. For acclimatization after regeneration from the tolerant calli only five salt tolerant plantlets developed from Torpe variety and eight salt tolerant plantlets developed from Siberian variety that succeeded to complete their life cycle and reached maturity stage and produced seeds. These regenerated genotypes were referred as T1, T2, T3, T4 and T5, the five regenerated plants developed from Torpe variety that tolerate NaCl concentration up to 16000 ppm, as well as, S1, S2, S3, S4, S5, S6, S7 and S8, the eight regenerated plants developed from Siberian variety that tolerate NaCl concentration up to 12000 ppm. The molecular characterization of the four canola varieties and their newly developed salt tolerant genotypes were performed using RAPD and ISSR analyses.

Key Words: *Brassica napus*, canola, regeneration, *in vitro* selection, salt stress, RAPD, ISSR.

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LIST OF ABBREVIATION

2,4-D	2,4-Dichlorophenoxy acetic acid
BA	Benzyl Adenine
BAP	Benzyl Amino Purin
IAA	Indole Acetic Acid
NAA	Naphthalene Acetic Acid
AgNo3	Silver Nitrate
AVG	Aminoethoxyvinylglycine
ACC	1-Amino-Cyclopropane-1-Carboxylic acid
RFLP	Restriction Fragment Length Polymorphism
RAPD	Randomly Amplified Polymorphic DNA