CONSERVATION OF BARTAMODA AND SAKKOTY DRY DATE PALM CVS. GERMPLASM

By

AMAL ABD EL-LATIF EL-ASHRY ABD EL-MONAEM

B.Sc. Agric. Sc. (Pomology), AlexandriaUniversity, 1999
M.Sc. Agric. Sc. (Pomology), Ain Shams University, 2006

A thesis submitted in partial fulfillment
of
the requirements for the degree of

DOCTOR OF PHILOSOPHY
in
Agricultural Science
(Pomology)

Department of Horticulture
Faculty of Agriculture
Ain Shams University

2013

Approval Sheet

CONSERVATION OF BARTAMODA AND SAKKOTY DRY DATE PALM CVS. GERMPLASM

By

AMAL ABD EL-LATIF EL-ASHRY ABD EL-MONAEM

B.Sc. Agric. Sc. (Pomology), AlexandriaUniversity, 1999

M.Sc. Agric. Sc. (Pomology), AinShamsUniversity, 2006

This thesis for Ph.D. degree has been approved by:

Dr. El- SayedGomaa Ibrahim ……
Head of Research, Horticulture Research Institute, A. R. C.

Dr. Ibrahim Mohamed Dosouky ……
Prof. Emeritus of Pomology, Fac. of Agric., Ain Shams Univ.

Dr. Ahmed Abd El - Hamid Awad…….
Associate Prof. of Pomology, Fac. of Agric., Ain Shams Univ.

Dr. AssemDosoukyShaltout ……..
Prof. Emeritus of Pomology, Fac. of Agric., Ain Shams Univ.

Date of Examination: 8 /10 /2013

CONSERVATION OF BARTAMODA AND SAKKOTY DRY DATE PALM CVS. GERMPLASM

By

AMAL ABD EL-LATIF EL-ASHRY ABD EL-MONAEM

B.Sc. Agric. Sc. (Pomology), AlexandriaUniversity, 1999

M.Sc. Agric. Sc. (Pomology), Ain – ShamsUniversity, 2006

Under the supervision of:

Dr. AssemDosoukyShaltout

Prof. of Pomology, Dept. of Horticulture, Faculty of Agriculture, AinShamsUniversity (Principal Supervisor)

Dr. Ahmed Abd El - Hamid Awad

Associate Prof. of Pomology, Dept. of Horticulture, Faculty of Agriculture, AinShamsUniversity

Dr. Mohamed Kamal El - Bahr

Research Prof. of Plant Biotechnology, NationalResearchCenter

ABSTRACT

AmalAbd El-Latif El-Ashry. Conservation of Bartamoda and Sakkoty Dry Date Palm cvs. Germplasm. Ph.D Thesis, Department of Horticulture, Faculty of Agriculture, University of Ain Shams, 2013.

The aim of this study was to establish an applicable method for *in vitro* preservation of two Egyptian dry date palm cultivars i.e., Bartamod and Sakkoty using slow growth and cryopreservation techniques. With maintaining the viability and the potential of regeneration during and after *in vitro* preservation. The molecular characterization of the preserved cultures for these two cultivars was performed to investigate the genetic stability of the preserved cultures using Random Amplified Polymorphic DNA (RAPD) technique. *In vitro* cultures of those two cultivars were obtained by culturing shoot tips excised from off-shoots. Embryogenic cultures were proliferated on MS medium supplemented with 10 mg/ l 2, 4- D+ 3 mg/l 2iP. For low temperature preservation, cultures of the two cultivars were incubated at 5°C for twelve months. Generally, survival percentage was decreased as storage period increased in both two cultivars. Storing at low temperature (5 ° C) increased the survival percentage and subsequently decreased the mortality percentage of embryogenic callus cultures of the two cultivars. After twelve months Bartamodaregegistred 97.7 % and Sakkoty registered 88. 8 %of survival with considerable browning specially with Bartamoda cultivar. At low temperature, rate of the germinated embryos of the two cultivars was increased as increasing of storage period. Also, the effect of preservation in complete dark at room temperature conditions was investigated.Storing the embryogenic callus cultures of the two cultivars in dark condition significantly increased the survival percentage compared with those stored in light. It was found that survival percentage significantly declined after twelve months of preservation. Sakkoty registered high percentage of survival compared with Bartamoda cultivar. It was found that dark condition had negative effects on germination of embryogenic cultures in both two cultivars of date palm. The role of mannitol, sorbitol and sucrose as osmotic agents was examined. Three concentrations of each one were tested (20, 40 and 60 g/l). It was found adding 40 or 60 g/ l mannitol or 20 g/ l sorbitol gave the highest survival percentage for Bartamoda cultivar. As for Sakkoty cultivar the three concentrations of sucrose gave the highest survival percentage. As for cryopreservation techniques, it was found that adding 0.5 or 0.7 M sucrose to the pretreatment medium gave moderate survival

percentage for embryos and callus of Sakkoty and for Bartamoda embryos. Exposing the explants to desiccation in the air laminar flow for two hours increased the survival percentage and subsequently the recovery percentages. Using encapsulation technique increased the survival percentage and subsequently the recovery percentage. The highest survival and recovery percentages were recorded with adding 0.5 and 0.7 M sucrose in the pretreatment medium and encapsulated with 4, 5 % sodium alginate for embryos and callus. Bartamoda recovery percentage was low it does not exceed 10 %. Genetic stability was tested using RAPD - PCR analysis. PCR products revealed that at DNA molecular level and RAPD analysis of *in vitro* preservation treatments of the two date palm cultivars exhibited genetic variations.

Key Words: Date palm, Bartamoda , Sakkoty*in vitro* preservation, low temperature, darkness, osmotic agents, cryopreservation, RAPD analysis.

ACKNOWLEDGMENT

I wish to offer, my sincere gratitude to Prof. Dr. AssemShaltout Professor of Pomology , Hort., Dept., Fac. of Agric., Ain Shams Univ., for his supervision, guidance, encouragement and his endless advice during the whole work.

I am thankful to Dr. Ahmed Abd El - Hamid, Associate Professor of Pomology, Hort., Dept., Fac. of Agric., Ain Shams Univ., for his supervision, guidance, endless advice and help in the preparation of the manuscript.

Great thanks are offered to Prof. Dr. Mohamed kamal El- Bahr Professor of Plant Biotechnology N. R. C. for his supervision, suggestions and constructive criticism during the whole work.

Many thanks are offered to Prof. Dr. ShawkyAbd El- Hamid Bekheet Professor of Plant Biotechnology N RCfor his guidance, encouragement, suggestions and helping in preparing the manuscript.

Also, many thanks are offered to Dr. Mohammad Mattar Associate professor of Plant Biotechnology NRC for his encouragement, suggestions and helping during the whole work.

I wish to offer my great gratitude to Dr. Ahmed MagdyGabr Researcher of Plant Biotechnology NRCfor his guidance, endless advice and encouragement during the whole work.

I wish to offer many thanks for all of my family who stood beside me, encouraged and helped me to complete this work.

CONTENTS

LIST OF TABLES