

**USE OF TISSUE CULTURE IN THE PRODUCTION
OF SECONDARY METABOLITES FROM
CHICORY (*Cichorium intybus*) PLANT**

By

MARWA OTHMAN MOHAMED SAYED

B.Sc. Agric. Sc. (Plant production), Faculty of Agric., Ain Shams University, 2014

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This Thesis for M.Sc. Degree has been approved by

Dr. Hekmat Yehia Ahmed Massoud

Professor Emeritus of Ornamental and Medicinal Plants, Faculty of
Agriculture, Mansoura University.

Dr. Asmaa Mohammed Mohammed Abd El-Gyed

Associate Professor of Ornamental, Medicinal and Aromatic Plants,
Faculty of Agriculture, Ain Shams University.

Dr. Abdelaziz Mohamed Hosni

Professor Emeritus of Ornamental, Medicinal and Aromatic Plants,
Faculty of Agriculture, Ain Shams University.

Dr. Laila Mohamed Helmi

Professor Emeritus of Ornamental, Medicinal and Aromatic Plants,
Faculty of Agriculture, Ain Shams University.

Date of Examination: 22/12/2019

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MARWA OTHMAN MOHAMED SAYED

B.Sc. Agric. Sc. (Plant production), Faculty of Agric., Ain Shams University, 2014

Under the Supervision of

Dr. Laila Mohamed Helmi

Professor Emeritus of Ornamental, Medicinal and Aromatic Plants,
Horticulture Department, Faculty of Agriculture, Ain Shams
University. (Principal Supervisor)

Dr. Abdelaziz Mohamed Hosni

Professor Emeritus of Ornamental, Medicinal and Aromatic Plants,
Horticulture Department, Faculty of Agriculture, Ain Shams
University.

ABSTRACT

Marwa Othman Mohamed Sayed: Use of Tissue Culture in the Production of Secondary Metabolites from Chicory (*Cichorium intybus*) Plant. Unpublished M.Sc. Thesis, Horticulture Department, Faculty of Agriculture, Ain Shams University, 2020.

All experiments of this research study were carried out in the plant tissue culture laboratory of the Agricultural Botany Department, Faculty of Agriculture, Ain Shams University, Shoubra El-kheima, Cairo, Egypt. Experiments on chicory plant were executed for the duration of two consecutive years 2017 and 2018. Chicory (*Cichorium intybus* L.), which belongs to Asteraceae family, is considered as an important medicinal plant due to the presence inside it of many bioactive substances such flavonoids, phenolic compounds, alkaloids, steroids, terpenoids, coumarines, cichoriin, esculetin, inulin, sesquiterpene lactones, chicoric acid, caffeic acid and some vitamins.

In this research, seed sterilization process aseptically was accomplished using three sterilizers namely mercuric chloride, Clorox and hydrogen peroxide in various concentrations and periods (0.1 % for 3-5 min., 10 % for 10 min and 12 % for 20 min), respectively. Clorox showed the best antiseptic effect with the lowest contamination percentages (2.5 % and 3.8 %). *In vitro* germination of chicory seeds used as initial explants were carried out using agar and half-strength Murashige and Skoog (MS) media to grow aseptic seedlings to provide for leaf explants from which callus is induced. Half-strength MS medium was the best in terms of the seedling growth rate (leaf number, width and length).

For friable callus induction, chicory leaf explants were abaxially inoculated on the surface of full-strength MS medium supplemented with different combinations of five plant growth regulators; NAA at 0.5 – 2.0 – 3.0 – 5.0 mg/l, IBA at 0.5 – 2.0 mg/l, 2,4-D at 3.0 – 5.0 mg/l, BA at 0.4 – 0.5 – 0.75 – 1.5 – 2.0 mg/l and IAA at 0.5 – 2.0 mg/l. Only MS medium

supplemented with NAA alone or combined with IBA produced the desirable friable callus and the optimum callus fresh weights were obtained with NAA at 3 mg/l plus IBA at 2 mg/l under total dark incubation condition.

Regarding callus content of phenolics, flavonoids and guaiacol peroxidase activity enzyme, NAA at 2 mg/l was the superior treatment. Meanwhile, reducing sugars, total free amino acids and polyphenol oxidase activity were increased with NAA at 5 mg/l plus IBA at 2 mg/l treatment. Moreover, the superior treatment for total soluble protein content and phenylalanine ammonia lyase activity was NAA at 3 mg/l plus IBA at 2 mg/l under total dark condition.

Keywords: *Cichorium intybus* L., Chicory, Asteraceae, Naphthalene acetic acid, NAA, Indole -3 -butyric acid, IBA, Auxin, Callus, *In vitro* culture, MS medium, Phenolic compounds, Flavonoids, Amino acids, Proteins, Reducing sugars, Enzyme activity, PAL, POD, PPO.

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