

SOME PHYSIOLOGICAL STUDIES ON STEVIA
(*Stevia rebaudiana*)

By

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ABSTRACT

An investigation was consummated at the Agricultural Botany, Plant Physiology Section, Faculty of Agriculture, Cairo University. Plant tissue culture experiments were carried out in Plant Physiology Research Laboratory, Agric. Bot. Dept., Fac. Agric., Cairo Univ., Giza Egypt. Acclimatization experiment was carried out in the GreenHouse of Olive Dept., Horticultural Institute, Agric. Res. Cent., Giza, Egypt. Anatomical studies were carried out in Research Park, Fac. Agric., Cairo Univ. Giza Egypt.

The current project was conducted during 2014 - 2018 to study the effect of Flurprimidol (Flur.), Paclobutrazol (PBZ) and thidiazuron (TDZ) on plantlets growth and acclimatization of *Stevia rebaudiana* Spanti and China-1 cultiv. Stem nodal segments containing axillary buds were used as an explant and cultured on MS medium containing 3% (w/v) sucrose and 0.8% (w/v) agar supplemented with various concentrations of Flur., PBZ and TDZ. In Spanti cultivar maximum number of branches (6.4) and (6.52) were obtained in MS medium supplemented with 0.12 ppm Flur and 0.2 ppm TDZ, respectively with an average of 56.4 and 36.66 leaves / plantlet, having an average shoot length of 4.18 and 3.22 cm, respectively. The best *in vitro* root induction (100%) was achieved on MS medium with 0.16 ppm Flur. with an average of 10 roots /plantlet and root length of 4.82 cm (Spanti cultivar). Furthermore, in China-1 cultivar MS medium supplemented with 0.16ppm Flur.induced the best morphological characteristics. As a result of anatomy, all studied growth regulators significantly enhanced the anatomical characters of stevia vars. leaf and stem. Flur.at 0.16mg\l surpassed, for instance, midvein and lamina thickness, length and width of leaf vascular bundle as well as stem diameter, xylem and phloem thickness. The rooted plantlets from explant planted on (MS) medium supplemented with 0.16ppm Flur. were successfully established in soil and grown to maturity at the survival rate of 100% in the greenhouse in (Spanti) and (China-1) cultivar Pot experiment was conducted in greenhouse on stevia plant to study the effect of both NH_4NO_3 and KNO_3 under the treatments of BA, Kin, and TDZ. NH_4NO_3 excelled KNO_3 fertilization on all growth characters except plant height. BA treatment tended to increase total fresh weight/ plant as compared to those of Kin and TDZ in some cases.

Molecular studies showed that Spanti and China-1 vars. are closely related with some slight differences. The protein bands which present in the treatments and absent in control might be responsible of increase the total chlorophyll a, chlorophyll b and total carotenoid as expressions of hormone affects, whereas Flur. and TDZ produced more greener plantlets in both vars. compared to PBZ (Yellowish plant). TDZ treatments induced the most of unique bands which may be explain the greatest number of branches and the highest percentage of callus which produced by using TDZ. The results indicated that treatments were successful in inducing further characterized for morphological and chemical composition traits.

Leaf chemical composition of N, P, K and protein as well as chlorophyll% were determined during 2017 and 2018 seasons. K concentration was higher under potassium nitrate fertilization when compared to ammonium nitrate fertilization. Potassium nitrate addition resulted in slight increase in chlorophyll a & b and carotenoids over ammonium nitrate addition.

Key words: Asteraceae, *Stevia rebaudiana*, *Stevia*, Tissue culture, Flurprimidol, PBZ, TDZ, Acclimatization, Ammonium nitrate, Potassium nitrate, Electrophoresis.

DEDICATION

I dedicate this work to whom my heartfelt thanks; to my late father's soul, my mother, my sisters and my brother for all the support they lovely offered to me along the period of my post-graduat studies and preparation of thesis desertation.

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God bless them all

ABBREVIATIONS

BA	: benzyl adenine
Bp	: Base pair
Car.	: Carotenoids
Chl.	: Chlorophyll
Chl.a	: Chlorophyll a
Chl.b	: Chlorophyll b
DAS	: Days After Sowing
Flur.	: Flurprimidol
FW	: Fresh weight
HPLC	: High Performance Liquid Chromatography
ISSR	: Inter-Simple Sequence Repeat
KDa	: Kilodalton
Kin.	: Kinetin
KNO₃	: Potassium nitrate
L	: liter
mg	: milligram
Min	: minute
MS media	: Murashige and Skoog
NH₄NO₃	: Ammonium nitrate
PBZ.	: Paclobutrazol
PPO	: Polyphenol oxidases
PX	: Peroxidase
TEMED	: Tetramethylethylenediamine
TDZ	: Thidiazuran
TLC	: Thin Layer Chromatography
TRT	: Treatment

CONTENTS

	Page
INTRODUCTION.....	1
REVIEW OF LITERATURE.....	5
1. Morphological studies.....	5
a. Stevia seed germination.....	5
b. Establishment and multiplication stages.....	9
(1) Surface sterilization.....	9
(2) Plant regeneration (The influence of plant growth regulators).....	11
(3) The effect of growth retardants and TDZ on morphological characteristics.....	17
(4) Rooting.....	24
c. Effect of plant growth retardants and TDZ on anatomy..	26
d. Effect of plant growth retardants and TDZ on acclimatization.....	29
e. Effect of fertilization (nitrogen source) and BA, Kin and TDZ on morphological characteristics in pot experiment....	33
2. Chemical composition.....	35
a. Effect of plant growth retardants and TDZ on protein profile in plantlets.....	35
b. Effect of plant growth retardants and TDZ on isoenzyme in plantlets.....	36
c. Effect of plant growth retardants and TDZ on chlorophyll in plantlets.....	40
d. Effect of fertilization (nitrogen source) and BA, Kin and TDZ on chlorophyll	43
e. Effect of fertilization (nitrogen source) and BA, Kin and TDZ on (NPK and total protein) in pot experiment.....	44

3. Molecular studies	47
MATERIALS AND METHODS	54
1. Morphological studies	54
a. Seed germination.....	54
b. Plant tissue culture studies.....	55
(1) Surface-sterilization.....	55
(2) Establishment and multiplication stages.....	56
(3) Effect of plant growth retardants and TDZ.....	57
(4) Anatomical study.....	58
c. Acclimatization.....	59
d. Pot experiment.....	60
2. Chemical composition	64
a. Leaf protein Electrophoresis.....	64
b. Isozymes electrophoresis (SDS- free).....	67
(1) Peroxidase (PX).....	69
(2) Polyphenol Oxidase (PPO).....	69
c. Plant pigments.....	70
d. Mineral concentration.....	71
(1) Digestion for total nitrogen %.....	71
(2) Digestion for total phosphorus and potassium % (dry ashing).....	72
(a) Phosphorus %.....	72
(b) Potassium %.....	73
3. Molecular studies	73

a. DNA isolation procedure.....	73
b. Polymerase chain reaction (PCR).....	75
4. Statistical analysis.....	77
RESULTS AND DISCUSSION.....	78
1. Morphological studies.....	78
a. Steiva seed germination.....	78
b. plant tissue culture studies.....	79
(1) Surface sterilization.....	79
(2) Establishment and multiplication effects of some growth regulators on morphological criteria.....	81
(3) Effects of growth retardants and TDZ on morphological characters.....	87
c. Effects of growth retardants and TDZ on anatomical studies through plant produced form tissue culture.....	100
(1) The Leaf.....	100
(2) The stem.....	105
d. Effects of growth retardants and TDZ in stevia on acclimatization through plant produced from tissue culture.....	109
e. Effects of both nitrogen sources and some growth regulators on plant growth characters through plant produced from pot experiment.....	112
2. Chemical composition.....	124
a. Effects of growth retardants and TDZ on biochemical identification in plant tissue culture experiment.....	124
(1) SDS- PAGE protein banding patterns of stevia leaves.....	124
(2) Isozymes banding patterns.....	128

(a) Peroxidase oxidase banding patterns.....	128
(b) Polyphenyl oxidase banding patterns.....	131
b. Effects of growth retardants and TDZ on stevia leaves plant pigments concentration on plants produced from tissue culture.....	134
c. Effects of both nitrogen sources and some growth regulators on stevia leaf pigments in plants from pot experment.....	140
d. Effects of both nitrogen sources and some growth regulators on chemical composition (NPK and total protein).....	146
3. Molecular studies (linkage Map between two stevia cultivars).....	149
a. Randomly Amplified Polymorphic DNA (RAPD) markers.....	149
b. Inter Simple Sequence Repeats (ISSRs) markers.....	155
CONCLUSION.....	162
SUMMARY.....	164
REFERENCES	174
APPENDIX.....	211
ARABICSUMMARY	

LIST OF TABLES

No.	Title	Page
1.	Some physical and chemical properties of the experimental soils..	61
2.	The chemical analysis of the used irrigation water.....	62
3.	Composition of resolving and stacking gels.....	65
4.	List of the primer names and their nucleotide sequences of RAPD and ISSR procedures used in the study.....	76
5.	. Effect of surface sterilization of stevia (<i>Stevia rebaudiana</i>) stem segments with different concentrations of sodium hypochlorite on survival, and some growth characters after six weeks.....	80
6.	Effect of different growth regulators treatment on growth characters in stevia (<i>Stevia rebaudiana</i> , Spanti cultivar) plant cultured on MS medium for six week.....	83
7.	Effect of different growth regulators treatment on growth characters in stevia (<i>Stevia rebaudiana</i> , China-1 cultivar) plant cultured on MS medium for six week.....	84
8.	Effects of Flur., PBZ and TDZ at different concentrations on average of data for morphological characteristics of <i>stevia rebaudiana</i> Spanti cultivar at first subculture.....	89
9.	Effects of Flur., PBZ and TDZ at different concentrations on average of data for morphological characteristics of <i>stevia rebaudiana</i> China-1 cultivar at first subculture.....	90
10.	Effects of Flur., PBZ and TDZ at different concentrations on average of data for morphological characteristics under study of <i>Stevia rebaudiana</i> Spanti cultivar at second subculture.....	92
11.	Effects of Flur., PBZ and TDZ at different concentrations on average of data for morphological characteristics under study of <i>Stevia rebaudiana</i> China-1 cultivar at second subculture.....	93
12.	Effects of Flur., PBZ and TDZ at different concentrations on average of data for morphological characteristics under study of <i>Stevia rebaudiana</i> plant Spanti cultivar at third subculture.....	95
13.	Effects of Flur., PBZ and TDZ at different concentrations on	96

	average of data for morphological characteristics under study of <i>Stevia rebaudiana</i> plant China-1 cultivar at third subculture.....	
14.	Microscopical measurements (μ) and counts of certain anatomical features in transverse sections through the Leaf of stevia plant treated with Flur. (0.16 ppm), TDZ (0.2 ppm) and PBZ (1.0 ppm) during 2016-2017.....	106
15.	Microscopical measurements (μ) and counts of certain anatomical features in transverse sections through the stem of stevia plant treated Flur. (0.16 ppm), TDZ (0.2 ppm) and PBZ (1.0 ppm) during 2016-2017.....	108
16.	Survival percentage, plant height and no. of leaves/plant during acclimatization as affected by Flur., PBZ and TDZ at different concentrations of <i>Stevia rebaudiana</i> spanti cultivar.....	109
17.	Survival percentage, plant height and no. of leaves/plant during acclimatization as affected by Flur., PBZ and TDZ at different concentrations of <i>Stevia rebaudiana</i> China-1 cultivar.....	110
18.	Effect of nitrogen sources and some plant growth regulators on morphological characteristics of <i>Stevia rebaudiana</i> Spanti cultivar plant at the first cut in the first season (2017)	114
19.	Effect of nitrogen sources and some plant growth regulators on morphological characteristics of <i>Stevia rebaudiana</i> Spanti cultivar plant at the first cut in the second season (2018)	115
20.	Effect of nitrogen sources and some plant growth regulators on morphological characteristics of <i>Stevia rebaudiana</i> China-1 cultivar plant at the first cut in the first season (2017)	116
21.	Effect of nitrogen sources and some plant growth regulators on morphological characteristics of <i>Stevia rebaudiana</i> China-1 cultivar plant at the first cut in the second season (2018)	117
22.	Effect of nitrogen sources and some plant growth regulators on morphological characteristics of <i>stevia rebaudiana</i> Spanti cultivar plant at second cut in the first season (2017)	120
23.	Effect of nitrogen sources and some plant growth regulator on morphological characteristics of <i>Stevia rebaudiana</i> Spanti cultivar plant at the second cut in the second season (2018)	121
24.	Effect of nitrogen sources and some plant growth regulators on morphological characteristics of <i>Stevia rebaudiana</i> China-1 cultivar plant at second cut in the first season (2017)	122
25.	Effect of nitrogen sources and some plant growth regulator on morphological characteristics of <i>Stevia rebaudiana</i> China-1	123

	cultivar plant at the second cut in the second season (2018)	
26.	Effect of flur. and TDZ concentrations (ppm) on SDS-protein banding patterns of proteins leaf (KDa) for stevia Spanti cultivar as the presence (1) and absence (0) of bands.....	125
27.	Effect of flur. and TDZ concentrations (ppm) on SDS-protein banding patterns of proteins leaf (KDa) for stevia China-1 cultivar as the presence (1) and absence (0) of bands.....	126
28.	Effect of flur. and TDZ concentrations (ppm) on leaf peroxidase isozymes of stevia cultivar (Spanti) as the presence (1) and absence (0) of bands.....	130
29.	Effect of flur. and TDZ concentrations (ppm) on leaf peroxidase isozymes of stevia cultivar (China-1) as the presence (1) and absence (0) of bands.....	131
30.	Effect of flur. and TDZ (ppm) on leaf polyphenol oxidase isozymes of stevia cultivar (Spanti) as the presence (1) and absence (0) of bands.....	132
31.	Effect of flur. and TDZ (ppm) on leaf polyphenol oxidase isozymes of stevia cultivar (China-1) as the presence (1) and absence (0) of bands.....	133
32.	Effect of some growth regulators on stevia (<i>Stevia rebaudiana</i> Spanti cultivar) on leaf pigments concentration at the third subculture.....	135
33.	Effect of some growth regulators on stevia (<i>Stevia rebaudiana</i> China-1 cultivar) leaf pigments concentration at the third subculture.....	138
34.	Effect of chemical fertilization and some plant growth regulators on plant pigments concentration under study of <i>Stevia rebaudiana</i> cultivar Spanti plant at the second cut in the second season.....	141
35.	Effect of chemical fertilization and some plant growth regulators on plant pigments under study of <i>Stevia rebaudiana</i> cultivar China-1 plant at the second cut in the second season.....	144
36.	Effect of chemical fertilization and some plant growth regulators on nitrogen, phosphorus, potassium (percentages) and protein under study of <i>Stevia rebaudiana</i> cultivar Spanti plant at the second cut in the second season.....	146
37.	Effect of chemical fertilization and some plant growth regulators nitrogen, phosphorus, potassium (percentages) and protein under study of <i>Stevia rebaudiana</i> cultivar China-1 plant	147

at the second cut in the second season.....	
38. Data matrix illustrating the presence or absence of RAPD bands produced by primer OP-A02 in the two varieties of <i>Stevia rebaudiana</i>	151
39. Data matrix illustrating the presence or absence of RAPD bands produced by primer OP-A09 in the two varieties of <i>Stevia rebaudiana</i>	152
40. . Data matrix illustrating the presence or absence of RAPD bands produced by primer OP-B11 in the two varieties of <i>Stevia rebaudiana</i>	152
41. Data matrix illustrating the presence or absence of RAPD bands produced by primer OP-C04 in the two varieties of <i>Stevia rebaudiana</i>	153
42. Data matrix illustrating the presence or absence of RAPD bands produced by primer OP-D01 in the two varieties of <i>Stevia rebaudiana</i>	154
43. Data matrix illustrating the presence or absence of RAPD bands produced by primer OP-D09 in the two varieties of <i>Stevia rebaudiana</i>	155
44. Data matrix illustrating the presence or absence of ISSR bands produced by primer 44B in the two varieties of <i>Stevia rebaudiana</i>	157
45. Data matrix illustrating the presence or absence of ISSR bands produced by primer 98B in the two varieties of <i>Stevia rebaudiana</i>	158
46. Data matrix illustrating the presence or absence of ISSR bands produced by primer HB-10 in the two varieties of <i>Stevia rebaudiana</i>	158
47. Data matrix illustrating the presence or absence of ISSR bands produced by primer HB-12 in the two varieties of <i>Stevia rebaudiana</i>	159
48. Species-specific RAPD and ISSR markers for <i>Stevia rebaudiana</i> species genotypes.	161