Available Online at http://www.journalajst.com

ASIAN JOURNAL OF SCIENCE AND TECHNOLOGY

ISSN: 0976-3376

Asian Journal of Science and Technology
Vol. 11, Issue, 01, pp.10712-10721, January, 2020

RESEARCH ARTICLE

APPRAISING ELICITATION POTENCY BY ELECTRIC CURRENT CORPORATED WITH CHITOSAN AND NONO- SELENIUM TO BROOST QUALE- QUANTITATIVE ESSENTIAL OIL YIELD PRODUCTION IN THYME

[1,*]Tarek El- Sayed, S.A. [2]Bosila, H.A. and [1]El- Sayed, S.A.

[1]Radiobiological Dept. and Nuclear Research Center, Atomic Energy Authority, Cairo Egypt
[2]Horticulture Department Medicinal and Aromatic Plant Division, Faculty of Agricultures, Al – Azhar University, Cairo, Egypt

ARTICLE INFO

Article History:
Received 15th October, 2019
Received in revised form
09th November, 2019
Accepted 27th December, 2019
Published online 31st January, 2020

Key words:
Thyme, Aromatic Plats, Elicitation, Electric Current, Chitosan, Nano- Selenium, Essential oil.

ABSTRACT

Thymus valuaris seeds, were subjected to DC electric current (E_{1-4}; 0, 100, 200, 500mA) before planting at filed; the resultant 4 months old plants were vegitatively sprayed every two week, up tell harvesting, with chitosan (C_{1-4}; 3,5,10g/L), Nano selenium oxide (N_{1-4}; 50, 100, 150ppb) and incorporated (EC), (EN) application treatments. ANOVA statistical analysis for the recorded data revealed that; E_{2-4}. C_{2-4}. N_{2-4} achieved significant positive impact C > N > E whereas, integrated (EC), (EN) performed synergistic significant positive impacts EC > EN quant – qualitative essential oil production. Therefore, E,C ,N,EC,EN increased essential oil yield, Kg/ha up to 25,69,58,80,71% over control (228.599Kg/ha); respectively Also, owing to increment dry herb yield, t/ha up to 15,27,22,31,28% over control (9.630t/ha) respectively, raise up the essential oil content. g/kg up to (1st /2nd harvest) (9/11), (31/37), (28/34), (35/42), (32/38)% over that of control (24.85/20.81/g/kg), respectively; exceedingly, scale up essential oil main components% that including thymole carvacrol, p-cymene, - terpinine up to (1st /2nd harvest) (6.8/12.5), (11.1/18.5), (7.8/16.0), (12.2/20.6), (10.1/18.5)% over that of control (8489/77.90%), respectively wherefore the précis results, strongly manifested EC exceed EN can be considered realistic biotechnological oriental strategy for quale – quantitative improvement thyme essential oil production , Ander field agriculture conditions..

Citation: ***Tarek El- Sayed, S.A. Bosila, H.A. and El- Sayed, S.A.*** **2020.** "Appraising elicitation Potency by electric current corporated with chitosan and nono-Selenium to broost quale- quantitative essential oil yield production in thyme", *Asian Journal of Science and Technology*, 11, (01), 10712-10721.

INTRODUCTION

Medicinal and aromatic plants produce essential oil, (EOs) in the form secondary metabolites (SMs) which have a beneficial impact on human health (1,2). Essential oils (EOs) are mixture of volatile compounds, mostly terpenes and their oxygenated derivatives produced in small quantities as secondary metabolites by many plants knowen as aromatic or medicinal plants. Since early times EOs have been used for flavor and fragrance in perfumery, pharmaceutical, cosmetic and food industries and antiseptic, healing and therapeutic active ingredients in folk medicine and aromatherapy. These EOs are useful as flavors or aroma enhancer in cosmetics, food additives, soaps, plastics, and perfumes. Moreover, curiosity about EOs applications that act as antimicrobial agents because of the broad range of activities, natural, origins, and generally recognized as safe status of EOs.

Corresponding author: **Tarek El- Sayed, S.A.,**
Radiobiological Dept. and Nuclear Research Center, Atomic Energy Authority, Cairo Egypt.

Currently, EOs are frequently studied for their antimicrobial (3,4), antifungal, herbicidal and anti- preoperative effect in orgainc farming and medicinal practice(5), antiulcer (6); antihuman thick (7); antioxidant (4), anti- inflammatory (8); repellent insecticidal antifeedant (9), antiviral (10); ovicidal (11); anesthetic (12); mollusidal (13); immunodulatory (14), larvicidal (15) as well as for their use as food preservations (9). are biodegradable general recognized as safe (16), represent a powerful tool to reduce the environmental impact of fruit production (17). In an attempt to prevent human health and avoid food losses in the face of global growing food industries, synthetic preservatives were introduced in food industries (18). For example lipid peroxidase in stored food may lead to rancidity and reduction of food quality(19). The consumption of spoiled food can cause a wide spectrum of human diseases(Furthermore, the development of resistant microorganisms to synthetic preservatives is another threat facing the continuous used of these chemicals. The different EOs compositions and different mutal ratios of compounds contained in the EOs may exert significant effect on their biological efficacy and their components demonstrate antimicrobial, antifungal and food preservative (20,21). Consequently, renewed interest has been placed on the discovery and use of natural bioactive resources in medicinal and aromatic plants to control diseases, food spoilage microorganisms and antoxidation (22 -24). Thyme (*Thyme Valgaris* L.), Family Lamiaceae is herbaceous and perennial aromatic, plant commonly consumed as herbal infusion and as condiment and spice (25). It is one of the most cultivated herb of thymus genus, being used in food, pharmaceutical cosmetic industries (26), inhibiting crimination, antispasmodic, antitussive, expectorants, bactericidal, and astringent. Traditionally, the herb has been used for treatment of dyspepsia, chronic gastric and diseases of the upper reparatory tract (24) .T. Vulgaris exist EO of different chemical components, thymol and its isomers carvacrol are the typical major substances found in thyme essential oil (TEO), (27)). TEO often inhibited a hedger biological efficacy compared to EOs from other aromatic plants (28). This why TEO have found application in medicine, they are traditionally used in treating bruises, various types of dermatitis and rheumatic types of pain, reducing seborrhea, regenerating capillary glands, and improving the condition of the hair. On account of its exptorant, spasmodic and antiseptic properties, it is notably used to treat a variety of illnesses of respiratory tract, such as flu, cold, sinusitis, chronic and acute bronchitis, tuberculosis, calming convulsive cough and irritable and spasmodic caught; due to its stimulant properties. It also acts as a nervous tonic and is used in asthenic state (29,30). TEO, it effect was related to their components which phenolic structure like carvactral, eugenol and thymol which are highly active against pathogen and their efficacy is often due to the synergy of different chemical component (31) TEO also used; in food industry as food flevour**.**

and aroma, antioxidants or substances that extend shelf of foods, reducing post harvest loss and preserving the quality of fruits, replace synthetic preservatives, protective agent for wheat grain intended for sowing and food production, as preservative agent for agriculture products and repellent against insects as biobesticid, control post**-** harvest diseases, bioherbicidal (32, 21). TEOs composition of the thyme species were used as antibacterial, anti- fungal, anti- oxidant, phytotoxic (33-35). Elicitation with physical (36) abiotic (37) biotic (38,39) elicitors is one of few strategies that commercial application in the improvement of secondary metabolites (SMs) production from plants as well as cell and organ cultures(40,41). Elicitation can be, also considered potential strategy in plant protection and biological control against microbial diseases and insect infestation, through induced systemic resistance (ISR) by induction the formation and enhance accumulation of secondary metabolite phytoalexins (PAs) (42- 44).

Electricity is not sharply defined in the natural sciences, through pacific properties belong to the core area of electricity electric field for example, are coursed by electric changes and can occur, e.g. under height voltage line (45). Electricity also include the electric current, which is a flow of electric charge carried by moving electrons in conductors or semi conductor or by ions in an electrolyte and is measured in ampere, Am. (46). Direct current (DC) means that there is non directional flow of electric charge, is directed in only one direction. In contrast, the movement of electric charge periodically reversed in direction in alternating electric current (AC) systems. Extensive field trials have been carried indicated stimulated growth and improved yield (47). Also, electromagnetic field resulted in significant increase bioactive secondary metabolites (48,49). Pre- sowing electric current of seeds affected plant growth, yield due to its influence varios physiological and biochemical activities in plants (50 ,51). Also (50) referred that electricity might be considered as reliable toole to improve yield in various crops. This conclusion has been confirmed by (52) whom exceedingly clarified that electric current would appear to be a general elicitor of plant secondary metabolites and have potential application both basic and commercial research. The electric current enhanced the membrane permeability for Ca^{2+}, whereby a rapid inflex of Ca^{2+} increased the level of (Ca^{2+}). This can generate stress and triggen signal transduction and the activation of metabolic processes within the plants (53). (54) declared that electric currents caused the formation of reactive oxygen species (ROS), whereby phenolic compounds where accumulated in garden cress, singlet oxygen superoxide, hydrogen peroxide (H_2O_2) and hydroxyl radical belong to the family of reactive oxygen species (ROS). Whereby abiotic and biotic stressors elicit an excess of ROS. Oxidative stress can damage cellular structure and function in plant or cause cell death (55). To control ROS toxicity antioxidant enzymes such as ascorbic peroxidase, catalase, super oxide dismutase are produced to scavenge ROS under stress, condition (56, 57). However, non enzymatic reduction of ROS can be achieved by antioxidant metabolism, including B **-** carotene (58), and phenolic compounds (59), Tacopherol (60). Only one study was found dealing with gene expression analysis to reveal molecular mechanism of how electric current can enhance the carotenoids in tomato (61).

Chitosan, of the most preferred biopolymer (Polysaccharide) due to its biodegradability, antimicrobial and non – toxic properties as well as being an economical material produced from west resources such as seafood shells (62, 63). It has significant devise uses in several fields of life i.e. plant sciences(64, 65**)** and medical science **(**66). Besides agriculture, chitosan has a large set of applications such as food, cosmetics and biomedical industries (67,68). Chitosan, is largely used to biotic and abiotic stresses it has been a bio fungicide, bio bactericide and bio- viscid which spurs plant defense system of plants and vegetables (69, 65). Furthermore, the growing demands for food also stimulated the increased use of industrial. Fertilizers, having catastrophic effects on human health. Therefore, the use of chitosan as bio fertilizer is considered (70). Chitsan effects on plant growth, yield attributes and physiological activities (71,72). Chitosan effect on fungi (73), antibacterial (71, 74), nematocidal effects (75), as virucideal (76, 77). Chitosan, Shows strong resistance to microbial diseases and insecticidal against various plant pests (78; 79). Therefore it would be utilize as a bio- insecticide for horticulture crops. Moreover, chitosan play a key role in the regulation of stomatal apparture and reduced the rate of transpiration when the plant is going through drought stress phase (80) to reduce post- harvest losses (43). The use nanotechnology has environmental benefits as an interdisciplinary science, can use as a potful tool to improve the agriculture sector and in important cases as crop production, uses less pesticides, fertilizers antimicrobial to aintan crops for long period (81). The interaction of plant cell with nanoparticles resulted in modification of plant gene expression and effect plant growth and development (82,83).

Nano particles as a novel abiotic elicitor in plant biotechnology in order fore production of desired secondary metabolites (81,84,85). Different studies have been performed to define the optimum agriculture conditions and exogenous chemical treatment which may boost the production of these voluble EOS. in the interest of medicinal, industrial supplies (86, 87). The ultimate aim of the recent researches in this area has been development of alternative control strategies to reduce depending on synthetic fungicides, bacteriosides, viruside, pesticides as well as elicitation to upraise biomass production and quality. Aside as far to allowed literature, studies on mult repeating elicitation potency with integrated elicitors were scarce to be considered specially under field conditions, therefore, the present research has been conducted.

MATERIALS AND METHODS

Elicitors treatment preparation

- Thyme seeds, just before planting were filled in filter paper which submerged into electrically conductive solution (2% Nacl), then exposed to DC electric current at 0.100, 200, 500 mA for 30 seconds. (E_{1-4}, respectively).
- Chitosan; 3,5,10 g/L dissolved C_{1-4} respectively in 1% acetic acid under gentile heating.PH elicitor solutions were adjusted to 5.7 pH with 1% Nacl solution. Tween -20 0.01% V/V was added as surfactant.
- Nano- Selenium oxide (30nm) solution at 0,50,100,150ppb concentrations N_{1-4} respectively with 0.01% (V/V) solution of Tween-20.

Planting

At 1st (2018) march EC. treated seeds were sowed in trays contained soil, sand, beat mixed (1:1:1 ratio V/V) subsequently established in greenhouse. After 10 days seedlings were fertilized 1.5g.Zn, 0.49g.cu, 1.2g. Fe, 1.29g.B, 0.29g. Mo/20L water. Seedling 3 weeks- age were trans planted to the field (Sandy soil) in plots 4x7m consisted 10 rows 40 cm apart 7m long and 20 cm inter spacing (12.5 plant/m^2) to give target plant population 52500 plant per Fadden (125000 plant /ha).

Field experiment: The layout of the experiment was in complete look design for three replicates and 28 application treatment (0 control;E_{2-4} 100,200,500 mA C_{2-4} 3,5,10g/L;N_{2-4} 50,100,150ppb; E_{2-4} x $C_{2-4;}$ E_{2-4} x N_{2-4}) Irrigation and fustigation (30kgN, 30kgP_2O_2,120KgK_2O/h)were carried out through drip irrigation system plants were kept weed free hand hgeing. No pesticide, antifungal, antimicrobial was applied plant . 2 months old were foliarly sprayed 28 application treatment every 2 weeks pre-1st harvest at 1st June and pre 2nd harvest at 1st August. Herb/ cut was dried in shade for 2 weeks and weighed to get harp yield, t/ha, that converted to get dry herb yield, t/ha (DHY, t/ha).

Essential oil extraction: Fifty gram dry herb plants for such 28 application treatment for 1st and 2st cut, was subjected to hydro- distillation for 3h. to obtain the essential oil content, then TEO were dried using anhydrous sodium sulfate and kept in amber glasses sealed with Teflon septa at 4°C until analysis. Essential oil yield content (EOY,g/Kg) and essential oil yield Kg/ha (EOY,Kg/ha) were calculated by the following equations:

EOY,g/Kg = (Extracted EO,g / 50 ground sample) × 1000
Y,Kg/m^2= DHY,/m^2×EOY,Kg/m^2
Which converted to EOY,Kg/ha.

Essential oil components identification: GC- MS analysis of EO was carried out using Agilone 7890A gas chromatography (Agilent Technologies Palo Alto, CH, USA) equipped wit a RTX- SNS capillary column (30mX 0.32mm, film thickness 0.25m) oven temperature was initially kept at 40°C for 2min, and then raised at the rate of 5°C/ min up to 210°C. Injection and detector temperatures were set at 290 and 300°C respectively. Helium was used as a carier gas at a flow rate of 2ml/ min, and 0.1ml samples were injected manually in the spit mode. Peak area percentage were calculated as quantitative data. The gas chromatograph was coupled to an Agilent 5975C(Agilent Technologies', Palo Alto, CA, USA) mass selective detector. Mass spectra were recorded in Elmode, in the range of m/Z 35-500. The El- MS operating parameter were ionization voltage 70 eV; ion source temperature, 250°C. retention indices were calculated for all components using a homologous series of n- alkanes (C5-24) injected under the same conditions as described above. The identification of individual constituents was accomplished by comparison of their spectra. With those from a viable MS and by comparison of the their experimentally determined retention indices.

Statistical analysis: The obtained data were subjected to analysis of variance, asides the differences between mean application treatments were statistically tested by the calculated LSD at 1% level.

RESULTS AND DISCUSSION

Statistical analysis of ANOVA revealed that individual E,C,N applications as will as integrated EC, EN application actuated significantly on DHY, t/ ha, TEO, g/Kg, TEOY, Kg/ ha and total % of main components of EO where as, the differences between means applications were tested according to the calculated LSD at 1% level (Table 1, 2).

Dry herb yield, t/ha (DHY, t/ha): DHY, t/ ha at, 1st, 2nd and their summation, at control (6.980, 2.650, 9.680 t/ha, respectively) were significantly augumed due to E_{2-4} application up to (15, 12.7%), (13,11.5%), (15,14.6%) over that of control; C_{2-4} up to (20,25,28%), (17,21,23%), (19,24,27%), over control; N_{2-4} up to (18,21,23%), (15,19,21%), (17,20,22%) over control, respectively. (Table 1, Figure 1), the highest DHY, t/ha was C_4 (27%) >N_4 (22%) >E_2 (15%) Whereas, E_{2-4} C_{2-4}, 9 incorporated application treatments performed significant synergetic increment ranged up to (18-33%), (14-27%), (19-28%), over control, respectively, (Table 1, Figure 2). Also, E_{2-4} N_{2-4}, 9 in corporated application treatments actuated synergistic significant increment ranged up to (20-29%), (17-25%), (19-28%) over control respectively (Table 1, Figure 3). The highest DHY, t/ha was E_2C_4 (31%) exceed E_2N_4 (28%). DHY, t/ha, in response for (E) application was in line with that reported previonly (54, 52). whom declared that (E) Influence various physiological and biochemical activities in plant. Also in line C application (88,71).

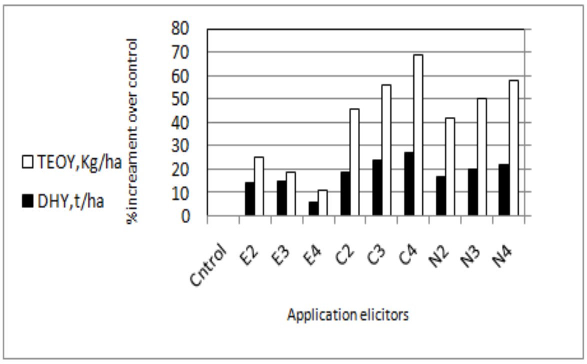


Figure 1. DHY, t/ha, TEOY, Kg/ha in response to E,C,N applications

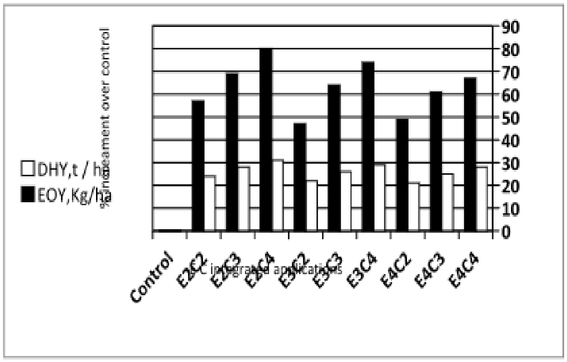


Figure 2. DHY, t/ha, TEOY, Kg/ha in response to EC applications

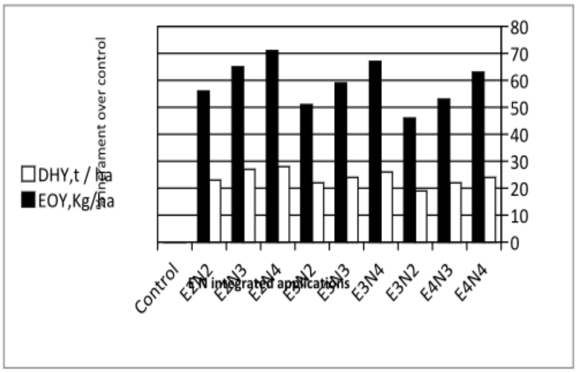


Figure 3. DHY,t/ha,TEOY,Kg/ha in response to EN applications

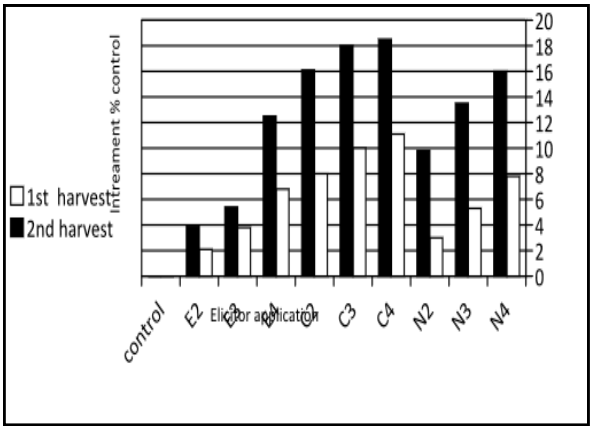


Figure 4. Total % TEO main components in response to E,C,N applications

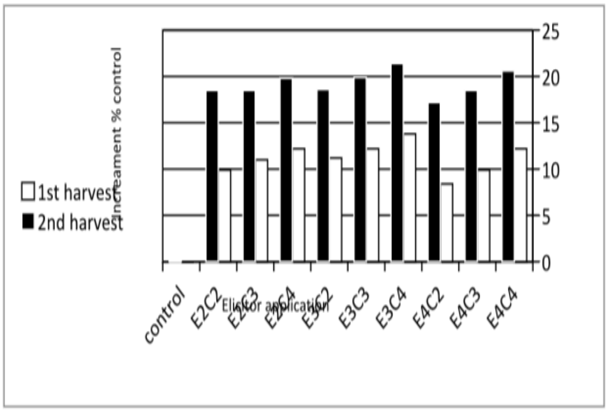
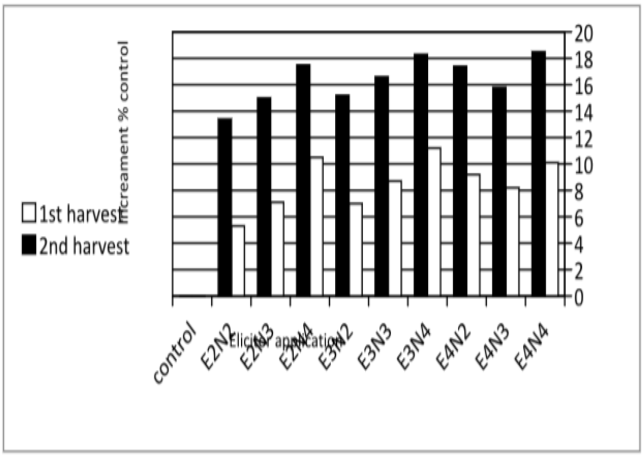


Figure 5. Total % TEO main components in response to EC applications



Aside C is considered as biofangicide, bioviruside, biobacteriocide, biopesticide which spurs plant defense system of plant (65, 75-77,79,65), and biofertilizer. C. Biofertilizers (70). N: application was in agreement with that has been reported by (83, 82)), Whom, declared that N modified of plant gene expression and affect plant growth and development. Also elicitation, can be considered potential strategy in plant protection and biological control against microbial diseases and insect infestation through induced systemic resistance (ISR) and initiation formation and accumulation (43, 89,90). These biosynthesized as a defense strategy by plants in response to perturbations under environmental condition (91). Also, elicitation enhancing formation and accumulation of bioactive secondary metabolites which play as a major role in adaptation of plant to the changes environment and over coming stress (34, 91).

Thyme essential oil herb content, g/kg (TEOC, g/kg): TEOC, g/Kg herb for control application treatment; 24.85, 20.81 at 1st and 2nd harvest, respectively; were significantly broost over control, E_{2-4} application up to (9,6,4%), (11,7,5%) respectively; C2-4 applications up to (21,25,31%), (15,30,37%), respectively; N2-4 applications up to (20,23,28%), (23,28,34%) over control respectively. Whereas, E_{2-4} C_{2-4}, 9 corporated application treatments achieved synergistic significant increment ranged (22-35%), (26-42%).Also, $E_{2-4}N_{2-4}$ (9wlegrated application treatment) activated significant synergistic increase ranged (21-38%) over control, respectively. The highest TEOC for E_2C (25,38%), exceed E_2N_4 (32,38%) over control, respectively (Table 1).

Table 1. Electrecity E), chitosan(C) and nano-selenium particles (N) as elicitors field application to attest their significant impact on thymeus herp and EO production

Elictor application	st harvest1			nd harvest2			Total 1st,2nd	
	DHY,t/ha	EOY,g./Kg	EOY,Kg/ha	DHY,t/ha	EOY,g./Kg	EOY,Kg/ha	DHY,t/ha	EOY,g./Kg
$E_1C_1N_1$	(0)6.980	(0)24.85	(0)173.453	(0)2.650	(0)20.81	(0)55.146	(0)9.630	(0)228.599
E2	(15) 8.027	(9)27.09	(25)217.452	(13)2.995	(11)23.05	(25)69.035	(15)11.022	(25)286.487
3	(12)7.818	(6)26.34	(19)205.926	(11)2.942	(7)22.30	(19)65.607	(14)10.760	(19)271.533
4	(07)7.469	(4)25.84	(11)192.998	(05)2.783	(5)21.80	(10)60.669	(06)10.252	(11)253.667
C2	(20)8.376	(21)30.07	(45)251.866	(17)3.101	(15)26.03	(64)80.719	(19)11.477	(46)332.585
3	(25)8.725	(25)31.06	(56)270.998	(21)3.207	(30)27.02	(57)86.653	(24)11.932	(56)357.651
4	(28)8.934	(31)32.55	(68)290.802	(23)3.260	(37)28.51	(69)92.943	(27)12.194	(69)383.745
N2	(18)8.236	(20)29.82	(42)245.597	(15)3.048	(23)25.78	(42)78.578	(17)11.284	(42)324.175
3	(21)8.446	(23)30.57	(49)258.194	(19)3.154	(28)26.53	(52)83.676	(20)11.600	(50)341.870
4	(23)8.586	(28)31.81	(58)273.121	(21)3.207	(34)27.77	(61)89.058	(22)11.793	(58)362.179
E2C2	(25)8.725	(25)31.06	(56)270.998	(23)3.260	(30)27.02	(60)88.085	(24)11.985	(57)359.083
2 3	(30)9.074	(30)32.31	(69)293.181	(24)3.286	(36)28.27	(68)92.895	(28)12.360	(69)386.076
42	(33)9.284	(35)33.55	(80)311.478	(27)3.366	(42)29.51	(80)99.331	(31)12.650	(80)410.809
E3C2	(18)8.236	(23)30.57	(45)251.775	(20)3.180	(28)26.53	(53)84.365	(22)11.416	(47)336.140
33	(28)8.934	((28)31.81	(64)284.191	(22)3.233	(34)27.77	(63)89.780	(26)12.167	(64)373.971
43	(31)9.144	(33)33.05	(74)302.209	(24)3.286	(40)29.01	(73)95.327	(29)12.430	(74)397.536
E4C2	(22)8.516	(22)30.32	(49)258.205	(19)3.154	(26)26.28	(50)82.887	(21)11.670	(49)341.092
34	(27)8.865	(27)31.56	(61)279.779	(14)3.207	(32)27.52	(60)88.256	(25)12.072	(61)368.035
44	(30)9.074	(30)32.31	(69)293.181	(24)3.286	(36)28.27	(68)92.895	(28)12.360	(67)386.076
E2N2	(25)8.725	(25)31.06	(56)270.998	(19)3.154	(30)27.02	(55)85.222	(23)11.879	(56)356.220
32	(28)8.934	(29)32.06	(65)286.424	(23)3.260	(35)28.02	(66)91.345	(27)12.194	(65)377.769
42	(29)9.002	(32)32.80	(70)295.331	(25)3.313	(38)28.76	(73)95.282	(28)12.317	(71)390.613
E3N2	(23)8.586	(23)30.57	(51)262.474	(18)3.127	(28)26.53	(50)82.959	(22)11.713	(51)345.433
33	(25)8.725	(27)31.56	(59)275.361	(21)3.207	(32)27.52	(60)88.257	(24)11.932	(59)363.618
43	(27)8.865	(31)32.55	(66)288.556	(23)3.260	(37)28.51	(69)92.943	(26)12.125	(67)381.499
E4N2	(20)8.376	(21)30.07	(45)251.866	(17)3.101	(25)26.03	(46)80.719	(19)11.477	(46)332.585
34	(22)8.516	(25)31.06	(25)264.507	(20)3.180	(30)27.02	(56)85.924	(22)11.696	(53)350.431
44	(25)8.725	(30)32.31	(63)281.905	(22)3.233	(36)28.27	(66)91.397	(24)11.958	(63)373.302
%LSD 1	0.003	0.02	0.005	0.002	0.03	0.003	0.004	0.005

E2-4 were electricity,100,200,500 Am ,respectively . C2-4 were chitosan.3,5,10 g,/L , respectively. N2-4 were Nano- selenium oxide particles \,50,100,200 ppb, respectively. Values between parenthesis were% potential increase over control.

Thyme essential oil yield, Kg/ha (TEOY, Kg/ha): TEOY for 1st, 2nd and their summation, Kg/ha , at control were; 173. 453, 55.146, 228.599, respectively; potentially were increased significantly over control, due to E2-4 application treatment up to (25,19,11%), (25,19,10%) , (25,19,11%) respectively: Also, due to C_{2-4} applications (45,56, 68%), (64,57,69%),(46,56,69%) respectively; Exceedingly due to N_{2-4} application treatment up to(42,49,58%), (42,52,61%), (42,50,58%), respectively (Table 1, Figure 1) . Therefore, TEOY in response of C > N >E and the highest increment for E2 (25%), C_4 (68%), N_4 (58%).potential.

$E_{2-4}C_{2-4}$, 9 incorporated application treatments achieved synergistic significant appraise for TEOY for 1st, 2nd and their summation ranged up to (45-80%), (50-80%), (49-80%), respectively. Also(Table 1, Figure 2),Wherease, E_{2-4} N_{2-4} applications ranged (45-70%), (46-73%), (46-71%), respectively (Table 1, Figure 3). The highest TEOY were found due to E_2C_4 up to (80, 80, 80%) that excel E_2N_4 up to (70, 73, 71%)over control (Table 1, Figure 3) for 1st, 2nd and their summation, respectively. The potential significant increase TEOC, g/Kg and TEOY, Kg/ ha were attributed to elicitors have a quantitative and qualitative impact SMs Essential oil phytoalexin, Phenolic group) which provide an important biological efficacy (29) through enhance the transcription of biosynthetic genes involved in SMs biosynthesis pathway. that led to enhanced formation and accumulation SMs (83, 38, 40, 92).

Qualitative characteristic for TEO: The analysis of 1st and 2nd harvest TEOY declared TEO contains 9 main components included 4 major components, thymol predominated follows P. cynene, carvactrol, - trepinine E2-4, C2-4, N2-4 at 1st and 2nd harvest, performed significant positive impacts, over % of control, for the total % of 9 TEO up to E (2.1, 3.8, 6.8%),(3.9,5.4,12.5), C (8.0,10.0,11.1%),(16.1,18.0,18.5%), N (3.0,5.3,7.8%), (9.8,13.5,16.0%), respectively (Table 2, Figure 4), while, E2-4 C2-4 and E2-4 N2-4 incorporated application treatment actuated synergistic significant increase, as % over control, ranged ((9.9-13.8%), (18.5-20.6%), ((15.3-11.3%) (13.4-18.5%), respectively (Table 2, Figure5,6),

Aside EC corborated application treatments at both 1st and 2nd harves, exceed EN. TEO often inhibited a high biological efficacy compared to EO from other aromatic plants (93). Also, its effects was related to their components, thymol, carvacole which high are highly active against pathogens and their efficacy is often due to the synergy of different chemical compounds (31). Since, any potential alterations of biological efficacy of EO should be verified given the well kmown fact that different EO compositions and different ratios of compounds contained in EO may exert on their biological efficacy (94). highlights, the effect of EC that exceed EN on overall metabolic processes in thyme plants and on their biological efficacy.

Table 2. Main coponents of thyme essential oil(TEO)extracted from thyme plants elicited with E,C,N, L and intractions EC,EN for 1st and2nd Thme havest

Monoterpene hidrocabons				Oxygenacted monoterpenes				Sasquetepene	Total %	Increament %0ver control
∞.pinene st2 st1	Myrecene st2 st1	-Terpinine st2 st1	P. cymenc st2 st1	Linolool st2 st1	Borneol st2 st1	Thymol st2 st1	Carvacro st2 st1	Tranc-Caryo-phlene 1st2 st	nd2 st1	ST 2ND1
0.51 –0.49	0.40 –0.59	4.65 –5.86	28.88- 27.60	0.33 –0.25	0.27 –0.35	35.42 -38.65	7.68 -9.85	1.25 1.95 -	77.90- 84.89	0 -0
0.45 –0.40	0.35 –0.47	4.80 -5.94	26.56-27.95	0.30 –0.22	0.25 –0.32	38.40 –40.13	7.92 -9.98	1.20 1.92 –	80.95 –86.61	3.9 –2.1
0.36 –0.41	0.34 –0.43	4.92 –6.27	27.22- 28.60	0.21 –0.23	0.30 –0.33	37.31 –42.67	8.09 –10.21	1.17 2.05 –	82.08 –88.14	5.4 –3.8
0.38 –0.42	0.30 –0.41	5.22 –6.21	28.88- 29.25	0.20 –0.21	0.28 –0.32	39.58 –40.95	9.61 -12.34	1.35 2.18 –	87.63 –90.64	12.5 –6.8
0.31 –0.35	0.31 –0.40	5.40 –6.35	29.88- 29.99	0.17 –0.20	0.25 –0.30	40.95 –41.86	10.91- 11.67	1.38 2.25 –	90.43 –92.50	16.1 –8.0
0.30 –0.33	0.32 –0.42	5.46 –7.42	30.21 -30.23	0.19 –0.21	0.26 –0.28	41.41 –42.32	11.01- 10.79	1.39 2.75 –	90.91 –93.39	18.0 –10.0
0.35 –0.32	0.31 –0.40	5.52 –6.49	30.54 -31.55	0.17 –0.19	0.24 –0.27	42.86 –42.77	10.11- 10.90	1.41 2.30 –	92.30 –94.30	18.5 –11.1
0.27 –0.30	0.30 –0.41	5.10 –6.00	28.22 -28.28	0.18 –0.20	0.26 –0.28	38.68 –40.59	10.42- 10.09	1.31 2.12 –	85.55 –87.46	9.8 –3.0
0.26 –0.28	0.31 –0.40	5.28 –6.14	29.22 -28.93	0.16 –0.19	0.25 –0.27	40.04 –41.50	10.71 -10.32	1.34 2.20 –	88.43 –89.37	13.5 –5.3
0.24 –0.27	0.32 –0.41	5.40 –6.28	29.88 -29.58	0.17 –0.18	0.25 –0.26	41.95 -432.41	9.91 -10.56	1.37 2.25 –	90.37 –91.32	16.0 –7.8
0.25 –0.27	0.30 –0.42	5.52 –6.42	30.54 -30.23	0.18 –0.20	0.24 –0.25	41.86 –42.32	11.11 -11.79	1.40 2.30 –	92.30 –93.30	18.5 –9.9
0.26 –0.28	0.30 –0.41	5.52 –6.49	30.54 -31.55	0.19 –0.20	0.24 –0.24	42.86 –62.77	10.11 -10.90	1.41 2.30 –	92.32 –94.25	18.5 –11.0
0.23 –0.25	0.32 –0.40	5.58 –6.56	30.88 -30.87	0.18 –0.21	0.25 –0.25	43.32 –44.23	10.21 -11.02	1.43 2.33 –	93.30 –95.22	19.8 –12.2
0.25 –0.31	0.33 –0.41	5.52 –6.49	30.54 -30.55	0.73 –0.75	0.78 –0.35	42.86 –42.77	9.11 –10.90	1.41 2.30 –	92.42 –94.44	18.6 –11.2
0.38 –0.36	0.41 –0.44	5.58 –6.55	30.87 -30.88	1.11 –1.12	1.20 –1.23	42.31 –42.22	9.20 –11.02	1.43 2.32 –	93.38 –95.27	19.9 –12.2
0.45 –0.51	0.46 –0.48	5.64 –6.62	31.21 -31.20	1.15 –1.17	1.25 –1.30	42.77 –42.68	9.31 –11.14	1.50 2.35 –	94.59 –96.60	21.4 –13.8
0.41 –0.40	0.42 –0.45	5.40 –5.31	30.20 -29.75	1.17 –1.18	1.24 –1.31	41.20 –42.56	9.01 –10.67	1.38 2.27 –	91.32 –92.01	17.2 –8.4
0.45 –0.48	0.41 –0.47	5.52 –6.42	30.54 -30.22	0.62 –0.65	0.51 –0.55	42.86 –42.32	9.11 –10.79	1.40 2.30 –	92.32 –93.30	18.5 –9.9
0.41 –0.45	0.35 –0.45	5.58 –6.56	30.88 -30.87	0.45 –0.48	0.42 –0.44	43.32 –43.51	10.21 -11.02	1.42 2.33 –	93.95 –95.22	20.6 –12.2
0.20 –0.25	0.31 –0.40	6.28 –6.14	30.21 -28.92	0.18 –0.25	0.20 –0.29	40.04 –41.50	8.71 –10.32	1.34 2.20 –	88.33 –89.41	13.4 –5.3
0.27 –0.30	0.33 –0.45	6.34 –7.21	30.55 -30.25	0.30 –0.35	0.25 –0.31	40.50 –39.25	8.81 –11.44	1.35 2.22 –	89.57 –90.92	15.0 –7.1
0.25 –0.27	0.32 –0.35	6.46 –7.42	31.21 -31.23	0.32 –0.38	0.27 –0.42	41.41 –40.53	9.01 –11.79	1.40 2.28 –	91.53 –93.79	17.5 –10.5
0.34 –0.37	0.35 –0.40	6.34 –6.21	30.55 -30.25	0.35 –0.40	0.29 –0.45	40.50 –40.95	8.81 –10.44	1.35 2.23 –	89.76 –90.82	15.2 –7.0
0.38 –0.39	0.37 –0.41	5.40 –6.52	29.88 -30.90	0.37 –0.42	0.31 –0.46	41.95 –40.10	9.91 –11.67	1.38 2.25 –	90.82 –92.25	16.6 –8.7
0.34 –0.48	0.45 -0.55	5.52 –7.49	30.54 -31.55	0.48 –0.50	0.45 –0.52	41.86 –39.95	10.11 -11.90	1.41 2.30 –	92.14 –94.35	18.3 –11.2
0.53 –0.69	0.51 –0.62	5.46 –6.35	30.21 -29.90	0.55 –0.61	0.50 –0.55	41.41 –41.86	10.01 -10.76	1.38 2.28 –	91.46 –92.72	17.4 –9.2
0.60 –0.74	0.63 –0.71	5.40 –6.28	29.88 -29.58	0.62 –0.67	0.52 –0.58	41.41 –41.40	8.91 –10.56	1.36 2.25 –	90.22 –91.88	15.8 –8.2
0.52 –0.81	0.54 –0.65	5.52 –6.42	30.54 -30.23	0.50 –0.60	0.40 –0.45	42.86 –43.32	9.11 –10.79	1.40 2.30 –	92.30 –93.47	18.5 –10.1
-	-	0.02 –0.03	0.9 –0.11	-	-	0.06 –0.08	0.2 –0.03	-	-	-

E2-4 were electricity,100,200,500 Am ,respectively . C2-4 were chitosan.3,5,10 g,/L , respectively.N2-4 were Nano- selenium oxide particles,50,100,200 ppb, respectively.Values between parenthesis were % potential increase over control.

Conclusion

Elicitation, at field application have quanti- qualitative significant positive impact with C>N>E elicitors aside EC performed synergistic significant positive impact exceeded EN application. Nevertheless we are aware that this finding will have to be verified at more localities and in more aromatic plant species, not only to confirm this phenomenon, but also to generalize the same by comparing the results for multiple plant species. We are also aware that the performed elicitation may have qualitative and quantitative impact on other SMs to exert significant effects on their biological efficacy, highlights in will efficacy electric current integrated with biotic or abiotic elicitor might be considered as reliable oriental. Technological strategy for quali – quantitative improvement essential oil in medicinal and aromatic plants under field conditions.

REFERENCES

1-Roohinejad, S., Koubaa, M., Barba, F. J., Leong, S. Y., Khelfa, A., Greiner, R., & Chemat, F. 2017. Extraction methods of essential oils from herbs and spices. Essential oils in food processing (pp. 21–55).

2-Jasminka Giacometti, Danijela Bursać Kovačević, Predrag Putnik, Domagoj Gabrić, Tea BilušićGreta Krešićd, Višnja Stulić Extraction of bioactive compounds and essential oils from mediterranean herbs by conventional and green innovative techniques: A review Food Research International 113 (2018) 245–262

3-Abhay K.Pandey1, PradeepKumar2 PoojaSingh Nijendra N. Tripathi 1 and Vivek K. Bajpai Essential Oils: Sources of Antimicrobials and Food Preservatives in Microbiology,7:1-14(2017).

4-Bhat G, Rassol S, Rehman S, Ganaie M, Qazi PH, Shawl AS. 2016. Seasonal variation in chemical composition, antibacterial and antioxidant activities of the essential oil of leaves of Salvia officinalis (sage) from Kashmir, India. *J Essent Oil Bearing Plant.,*19:1129–40.

5-Ersilia Alexa Renata Maria Sumalan, Corina Danciu, Diana Obistioiu, Monica Negrea, Mariana-Atena Poiana, Cristian Rus, Isidora Radulov, Georgeta Pop and Cristina Dehelean, 2018.

6-Dordevic, S., Petrovic, S., Dobric, S., Milenkovic, M., Vucicevic, D., Zizic, S. et al. 2007. Antimicrobial, anti-inflammatory, anti-ulcer and antioxidanta ctivities of *Carlinaacanthifolia* root essential oil. *J. Ethnopharmacol.* 109,458–463. doi:10.1016/j.jep.2006.08.021

7-Inouye, S., Takizawa, T. and Yamaguchi, H. 2001. Antibacterial activity of essential oils and the irmajorconstituents agains trespiratory tract pathogens by gaseous contact. *J.Antimicrob.Chemother.* 47,565–573

8-Singh, S., Majumdar, D.K. and Rehan, H.M.S. 1996. Evaluation of anti-in flammatory potential offixedoilof *Ocimumsanctum* (Holybasil) and its possible mechanism of action. *J.Ethnopharmacol.* 54,19–26.

9-Pandey, A.K., Singh, P., Palni, U.T. and Tripathi, N.N. 2014. *Invivo* evaluation of two essential oil basedbotanical for mulations (EOBBF) for the useagainst storedproductpests, *Aspergillus* and *Callosobruchus* (:Bruchidae) species. *J.StoredProd.Res.* 59,285–291.

10-Maurya, S., Marimuthu, P., Singh, A., Rao, G.P. and Singh, G. 2005. Antiviralactivity of essential oils and acetone extracts of medicinal plants against papayaringspotvirus. *J.Essent.OilBear.Plants* 8,233–238.

11-Pandey, A.K. and Tripathi, N.N. 2011. Post-harvest fungal and insect deterioration of pigeon pea seeds and the management by plant volatiles. *J. Ind.Bot.Soc.* 90,326–331.

12-Ghelardini, C., Galeotti, N. and Mazzanti, G. 2001. Localanesthetic activity of mono terpenes and phenylpropanes of essential oils. *Planta Med.* 67,564–566. doi:10.1055/s-2001-16475.

13-Fico, G., Panizzi, L., Flamini, G., Braca, A., Morelli, I., Tome, F., et al. 2004. Biologicals creening of *Nigellaelamascena* for antimicrobial and molluscicidal activities. *Phytother. Res.* 18,468–470.doi:10.1002/ptr.1454.

14-Mediratta, P.K., Sharma, K.K. and Singh,S. 2002. Evaluation of immunomodul at ory potential of *Ocimumsanctum* seeds oil and its possible mechanism of action. *J.Ethnopharmacol.* 80,15–20.

15-Jantan, I., Ping, W.O., Visuvalingam, S.D. and Ahmad., N.W. 2003. Larvicidal activity of the essential oils and methan olicextractso fMalaysian plants on *Aedesaegypti*. *Pharma.Biol.* 41,234–236.

16-Sivakumar, D., Bautista-Baños, S., 2014. A review on the use of essential oils for postharvest decay control and maintenance of fruit quality during storage. Crop Protection 64, 27-37.

17-Burt, S. 2004. Essential oils: Their antibacterial properties and potential applications in foods—A review. *Int. J. Food Microbiol.,* 94, 223–253.

18-Tripathi, N.N. and Kumar, N. 2007. *Putranjivaroxburghii* oil-Apotential herb AL preservative for peanuts during storage. *J.StoredProd.Res.* 43,435–442

19-Dai J, Zhu L, Yang L, Qiu J. 2013. Chemical composition antioxidant and antimicrobial activities of essential oil from Wedelia prostrate. EXCLI J.

20-Sonker, N., Pandey, A.K. and Singh, P. 2015. Efficiency of *Artemisianilagirica* (Clarke)Pamp essential oilas amycotoxicant against post harvest mycobiota of tablegrapes. *J.Sci.FoodAgric.* 95,1932–1939.

21-Noha Khalil, Mostafa Fekry, Mokhtar Bishr, Soheir El-Zalabanib, Osama Salamaa Foliar spraying of salicylic acid induced accumulation of phenolics, increased radical scavenging activity and modified the composition of the essential oil of water stressed Thymus vulgaris Plant Physiology and Biochemistry 123 (2018) 65–74 L.

22-Sharifi-Rad J, Salehi B, Stojanović-Radić ZZ, Fokou PVT, Sharifi-Rad M, Mahady GB, et al. Medicinal plants used in the treatment of tuberculosisethnobotanical and ethnopharmacological approaches. Biotechnol Adv2017; S0734–9750(17)30077–0

23-Elansary HO, Szopa A, Kubica P, Ekiert H, Ali HM, Elshikh MS, et al. Bioactivities of traditional medicinal plants in Alexandria. Evid-Based Complementary Altern Med. 2018;2018:1463579.

24-Adrar N, Oukil N, Bedjou F. Antioxidant and antibacterial activities of Thymus numidicus and Salvia officinalis essential oils alone or in combination. Ind Crop Prod. 2016;88:112–9.

25-Helmy, W. A., Farrag, A. A., & Hasaballah, A. A. (2015). Chemical composition and biological activity of aqueous and methanolic extracts of thyme (Thymus vulgaris). International Journal of Advanced Research, 3, 1285e1305.

26-Gavaric_, N., Kladara, N., Misanb, A., Nikolicc, A., Samojlikd, I., Mimica-Dukice, N., et al. (2015). Postdistillation waste material of thyme (Thymus vulgaris

L., Lamiaceae) as a potential source of biologically active compounds. Industrial Crops and Products, 74, 457e464

27-Stahl-Biskup, E., Saez, F., 2002. Thyme: The Genus Thymus. CRC Press, Francisco Saez. Pavela, R., 2011. Insecticidal and repellent activity of selected essential oils against of the pollen beetle. Meligethes Aeneus 34, 888–892.

28-Matusinsky, P., Zouhar, M., Pavela, R., Novy, P., 2015. Antifungal effect of five essential oils against important pathogenic fungi of cereals. Ind. Crop. Prod. 67, 208–215.

29-Marinelli, O., Iannarelli, R., Morelli, M.B., Valisi, M., Nicotra, G., Amantini, C., Cardinali, C., Santoni, G., Maggi, F., Nabissi, M., 2016. Evaluations of thyme extract effects in human normal bronchial and tracheal epithelial cell lines and in human lung cancer cell line. Chem. Biol. Interact. 256, 125–133.

30-Nikolic_, M., Glamo_clija, J., Ferreira, I. C. F. R., Calhelha, R. C., Fernandes, A., Markovi_cc, T., et al. (2014). Chemical composition, antimicrobial, antioxidant and antitumor activity of Thymus serpyllum L., Thymus algeriensis Boiss. and Reut and Thymus vulgaris L. essential oils. Industrial Crops and Products, 52, 183e190.

31-Spadaro, D.; Gullino, M.L. Use of Essential Oils to Control Postharvest Rots on Pome and Stone Fruit. In Post-Harvest Pathology; Prusky, D., Gullino,M.L., Eds.; Springer International Publishing: Cham, Switzerland, 2014; pp. 101–110.

32-Tjaša Pršin, Sabina Anžlovar, Jasna Dolenc Koce The effect of thyme essential oil on germination and early growth of wheat Acta Biologica Slovenica (2018). Vol .61,St 1:3-12.

33-Kashkooli, A.B., Saharkhiz, M.J. (2014). Essential oil compositions and natural herbicide activity of four denaei Thyme (*Thymus daenensis* Celak.) ecotypes. J. Essent. Oil Bearing Plants. 17: 859-874.

34-Ozen, T., Demirtas, I. (2015). Antioxidative properties of *Thymus pseudopulegoides*: comparison of different extracts and essential oils. J. Essent. Oil Bearing Plants. 18: 496-506.

35-Chandra, M., Prakash, O., Bachheti, R.K., Kumar, M., Pant, A.K. (2016). Essential oil composition, phenolic constituents, antioxidant and pharmacological activities of *Thymus linearis* Benth. collected from Uttarakhand region of India. J. Essent. Oil Bearing Plants. 19: 277-289.

36-Dörnenburg,.H; Knorr, D. (1995).Strategies for the improvement of secondary metabolite production in plant cell cultures. *Enzyme Microb Technol. 17*, 674–684.

37-Radman, R.; Sae, T.; Bucke, C. and T. Keshavarz. (2003):Elicitation of plant and micropial systems Biotechnol. Appl Biocchem.37:91-102.

38-Randhir, R.; Lin, Y.-T.; Shetty, K. (2004). Stimulation of phenolics, antioxidant and antimicrobial activities in dark germinated mung bean sprouts in response to peptide and phytochemical elicitors. *Process Biochem. 39*, 637–646.

39-Angelova, Z.; Gergiev, S.; Roes, W. (2006). Elicitation of plants. *Biotechnol. l. Equip . 20*, 72–83.

40-Michał Świeca .(2016). elicitation and treatment with precursors of phenolics synthesis improve low-molecular antioxidants and antioxidant capacity of buckwheat sprouts Acta Sci. Pol. Technol. Aliment., 15(1), 17–28. DOI: 10.17306/J.AFS. 1.2.

41-Nieves Baenas, Cristina García-Viguera and Diego A. Moreno. (2014). Elicitation: A Tool for Enriching the Bioactive Composition of Foods *Molecules. 19*, 13541-13563.

42-DeWitte, A.; Leus, L.; Huylenbroeck, J-van; Bockstaele, E-van and M. Hofte (2007): Resistance reaction in rose leaves against powdery mildew (*Podosphaera pannosa*). Acta Horticulture; (751): 183-188.

43-Zheng Fangliang; Zheng WenWen; li liMei; Pan SiMing; Liu MeiChen; Zhang WenWen; Liu HongSheng; Zhu Chunyu.Chiosan controls postharvest decay and elicits defense response in kiwifruit. Food and Bioprocess Technology; 2017.10(11):1937-1945.

44-Thanh – Tam Ho; lee JongDu; Jeong CheolSeung; Paek Kee Yoeup; Park So Young. Improvement of biosynthesis and accumulation of bioactive compounds by elicitation in adventitious root cultures of polygonum multiflorum. Applied Microbiology and Biotechnology; 2018. 102(1):199-209.

45-Feynman, R.P., Leighton, R.B., Sands, M., 1964. The Feynman lectures on physics. Mainly Electromagnetism and Matter, vol. II Basic Books, New York

46-Horowitz, P., Hill, W., 2015. The Art of Electronics, 3 ed. Cambridge University Press, New York Dennis Dannehl Effects of electricity on plant responses Scientia Horticulturae 234 (2018) 382–392

47-Omran A.F. and A.M. Abdel-Wahab. (1974): Effect of treating cucumber seeds with electric current on growth and yield. Egypt.J. Hort 1(1): 79-88.

48-Hong-Ye.; Lin-Ling Haung; Shu- De Chen and Jian- Jiang Zhong (2004): Pulsed electric field stimulates plant secondary metabolism in suspension culture of Taxus chinensis. Biotechnol. Bioeng.; 88(6): 788-795.

49-Pietruszewski, S.; Muszynski, S. and A. Dziwulska. (2007): Electromagnetic field and electromagnetic radiation as non – invasive external stimulants for seeds (Selected methods and responses). International Agrophysics; 21(1): 95-100.

50-Bera, A. K.; Pati, M. K. and A. Bera. (2007): Electrotherapy of pre-sowing seeds: a novel technique for yield improvement in crop plants. (Indian Journal of Plant Physiology; 12(4): 301-311).

51-Bati, M.K.; Ghanti, P. and A.K. Bera. (2008): Effect of pre-sowing electric current treatment of seeds on growth and yield of bottle ground (Lagenaria siceraria Mol. Standl). (Research on Crops; 9(1): 79-81.

52-Kaimoyo, E.; Farag, M.A.; Sumer, L.W.; Wasmann, C.; Cuello, J.L. and H.Vanetten.(2008): Sub lethal levels of electric current elicit the biosynthesis of plant secondary metaboles. Biotechnology Progress; 24(2): 377-384.

53-White, P.J., Broadley, M.R., 2003. Calcium in plants. Ann. Bot. 92, 487–511.

54-Dannehl, D., Huyskens-Keil, S., Wendorf, D., Ulrichs, C., Schmidt, U., 2012. Influence of intermittent-direct-electric-current (IDC) on phytochemical compounds in garden cress during growth. Food Chem. 131, 239–246

55-Gaspar, T., Franck, T., Bisbis, B., Kevers, C., Jouve, L., Hausman, J.F., Dommes, J., 2002. Concepts in plant stress physiology. Application to plant tissue cultures. Plant Growth Regul. 37, 263–285.

56-Impa, S.M., Nadaradjan, S., Jagadish, S.V.K., 2012. Drought stress induced reactive oxygen species and anti-oxidants in plants. In: Ahmad, P., Prasad, M.N.V. (Eds.), Abiotic Stress Responses in Plants. Metabolism, Productivity and Sustainability. springer Science+Business Media, New York.

57-Zheng, G., Lv, H.P., Gao, S., Wang, S.R., 2010. Effects of cadmium on growth and antioxidant responses in Glycyrrhiza uralensis seedlings. Plant Soil Environ. 56, 508–515.
58-Penuelas, J., Munne-Bosch, S., 2005. Isoprenoids: an evolutionary pool for photoprotection. Trends Plant Sci. 10, 166–169
59-Kovacik, J., Klejdus, B., Backor, M., 2009. Nitric oxide signals ROS scavenger-mediated enhancement of PAL activity in nitrogen-deficient Matricaria chamomilla roots: side effects of scavengers. Free Radic. Biol. Med. 46, 1686–1693.
60-Krieger-Liszkay, A., Trebst, A., 2006. Tocopherol is the scavenger of singlet oxygen produced by the triplet states of chlorophyll in the PSII reaction centre. J. Exp. Bot. 57, 1677–1684
61-Dannehl, D., Huyskens-Keil, S., Eichholz, I., Ulrichs, C., Schmidt, U., 2011. Effects of direct-electric-current on secondary plant compounds and antioxidant activity in harvested tomato fruits (Solanum lycopersicon L.). Food Chem. 126, 157–165.
62-Dash, M.; Chiellini, F.; Ottenbrite, R.; Chiellini, E. Chitosan—A versatile semi-synthetic polymer in biomedical applications. Prog. Polym. Sci. 2011, 36, 981–1014
63-Shukla, S.K.; Mishra, A.K.; Arotiba, O.A.; Mamba, B.B. Chitosan-based nanomaterials: A state-of-the-art review. Int. J. Biol. Macromol. 2013, 59, 46–58.
64-Kaya, M.; Mujtaba, M.; Bulut, E.; Akyuz, B.; Zelencova, L.; Sofi, K. Fluctuation in physicochemical properties of chitins extracted from different body parts of honeybee. Carbohydr. Polym. 2015, 132, 9–16.
65-Kaya, M.; Akyuz, L.; Sargin, I.; Mujtaba, M.; Salaberria, A.M.; Labidi, J.; Cakmak, Y.S.; Koc, B.; Baran, T.; Ceter, T. Incorporation of sporopollenin enhances acid–base durability, hydrophobicity, and mechanical, antifungal and antioxidant properties of chitosan films. J. Ind. Eng. Chem. 2017, 47, 236–245.
66-Shamov, M.; Bratskaya, S.Y.; Avramenko, V. Interaction of carboxylic acids with chitosan: Effect of pK and hydrocarbon chain length. J. Colloid Interface Sci. 2002, 249, 316–321.
67-Mujtaba, M.; Salaberria, A.M.; Andres, M.A.; Kaya, M.; Gunyakti, A.; Labidi, J. Utilization of flax (Linum usitatissimum) cellulose nanocrystals as reinforcing material for chitosan films. Int. J. Biol. Macromol. 2017, 104, 944–952.
68-Akyuz, L.; Kaya, M.; Koc, B.; Mujtaba, M.; Ilk, S.; Labidi, J.; Salaberria, A.M.; Cakmak, Y.S.; Yildiz, A. Diatomite as a novel composite ingredient for chitosan film with enhanced physicochemical properties. Int. J. Biol. Macromol. 2017, 105, 1401–1411.
69-Li, B.; Shi, Y.; Shan, C.; Zhou, Q.; Ibrahim, M.; Wang, Y.; Wu, G.; Li, H.; Xie, G.; Sun, G. Effect of chitosan solution on the inhibition of Acidovorax citrulli causing bacterial fruit blotch of watermelon. J. Sci. Food Agric. 2013, 93, 1010–1015.
70-Agbodjato, N.A.; Noumavo, P.A.; Adjanohoun, A.; Agbessi, L.; Baba-Moussa, L. Synergistic effects of plant growth promoting rhizobacteria and chitosan on in vitro seeds germination, greenhouse growth, and nutrient uptake of maize (Zea mays L.). Biotechnol. Res. Int. 2016, 2016, 7830182.
71-Sathiyabama, M.; Akila, G.; Einstein Charles, R. Chitosan-induced defence responses in tomato plants against early blight disease caused by Alternaria solani (Ellis and Martin) Sorauer. Arch. Phytopathol. Plant Prot. 2014, 47, 1777–1787.
72-Zagzog, O.A.; Gad, M.M.; Hafez, N.K. Effect of Nano-chitosan on Vegetative Growth, Fruiting and Resistance of Malformation of Mango. Trends Hortic. Res. 2017, 6, 673–681.
73-Ben-Shalom, N.; Fallik, E. Further suppression of Botrytis cinerea disease in cucumber seedlings by chitosan-copper complex as compared with chitosan alone. Phytoparasitica 2003, 31, 99–102.
74-Algam, S.; Xie, G.; Li, B.; Yu, S.; Su, T.; Larsen, J. Effects of Paenibacillus strains and chitosan on plant growth promotion and control of Ralstonia wilt in tomato. J. Plant Pathol. 2010, 92, 593–600.
75-Escudero, N.; Lopez-Moya, F.; Ghahremani, Z.; Zavala-Gonzalez, E.A.; Alaguero-Cordovilla, A.; Ros-Ibañez, C.; Lacasa, A.; Sorribas, F.J.; Lopez-Llorca, L.V. Chitosan increases tomato root colonization by Pochonia chlamydosporia and their combination reduces root-knot nematode damage. Front. Plant Sci. 2017, 8.
76-Bondok, A. Response of Tomato Plants to Salicyli c Acid and Chitosan under Infection with Tomato mosaic virus. Am.-Eur. J. Agric. Environ. Sci. 2015, 15, 1520–1529.
77-Firmansyah, D. Use of Chitosan and Plant Growth Promoting Rhizobacteria to Control Squash Mosaic Virus on Cucumber Plants. Asian J. Plant Pathol. 2017, 11, 148–155.
78-Sahab, A.; Waly, A.; Sabbour, M.; Nawar, L.S. Synthesis, antifungal and insecticidal potential of Chitosan (CS)-g-poly (acrylic acid)(PAA) nanoparticles against some seed borne fungi and insects of soybean. Int. J. Chem. Tech. Res. 2015, 8, 589–598.
79-Li, Y.; Qin, Y.; Liu, S.; Xing, R.; Yu, H.; Li, K.; Li, P. Preparation, Characterization, and Insecticidal Activity of Avermectin-Grafted-Carboxymethyl Chitosan. BioMed Res. Int. 2016, 2016.
80-Lim, C.W.; Baek,W.; Jung, J.; Kim, J.-H.; Lee, S.C. Function of ABA in stomatal defense against biotic and drought stresses. Int. J. Mol. Sci. 2015, 16, 15251–15270.
81-Faezeh Ghanati1, Somayeh Bakhtiarian, Behrooz Mohammad Parast and Mahboobeh Keyhani Behrooz Production of New Active Phytocompounds by *Achillea millefolium* L. after Elicitation with Silver Nanoparticles and Methyl Jasmonate BIOSCIENCES BIOTECHNOLOGY RESEARCH ASIA (2014).
82-Remya Nair, Saino Hanna Varghese, Baiju G. Nair, T. Maekawa, Y. Yoshida, D. Sakthi Kumar Nanoparticulate material delivery to plants Plant Science 179 (2010) 154–163.
83-Sharifi-Rad1, M. Sharifi-Rad2, and J. A. Teixeira da Silva3Morphological, Physiological and Biochemical Responses of Crops (Zea mays L., Phaseolus vulgaris L.), Medicinal Plants (Hyssopus officinalis L., Nigella sativa L.), and Weeds (Amaranthus retroflexus L., Taraxacum officinal F. H. Wigg) Exposed to SiO2 NanoparticlesJ. Agr. Sci. Tech. (2016) Vol. 18: 1027-1040.
84-Hina Fazal Bilal Haider Abbasi Nisar Ahmad4 Mohammad Ali:(2016). Elicitation of Medicinally Important Antioxidant secondary Metabolites with Silver and Gold Nanoparticles in Callus Cultures of Prunella vulgaris L. Appl Biochem Biotechnol, 180:1076–1092.

85-Moharrami,F.; Hosseini, B.; Sharafi, A.; Farjaminezhad, M. Enhnced production of hyoscyamine and scopolamine from genetically transformed root culture of Hyoscyamus reticulatus L. elicited by iron oxide nanoparticles.In Vitro Cellulaar & Developmental Biology- plant;2017. 53(2):104-111.32.

86-Misra, N., Misra, R., Mariam, A., Yusuf, K., Yusuf, L., 2014. Salicylic acid alters antioxidant and phenolics metabolism in Catharanthus roseus grown under salinity stress. Afr. J. Tradit., Compl. Altern. Med. 11, 118–125.

87-Li, L., Dong, Y., Ren, H., Xue, Y., Meng, H., Li, M., 2017. Increased Antioxidant Activity and Polyphenol Metabolites in Methyl Jasmonate Treated Mung Bean (Vigna Radiata) Sprouts. Food Science and Technology, Campinas.

88-Reglinski, T.; Elmer, P.; Taylor, J.;Wood, P.; Hoyte, S. Inhibition of Botrytis cinerea growth and suppression of botrytis bunch rot in grapes using chitosan. Plant Pathol. 2010, 59, 882–890.

89-Zlotek, Affect of jasmonic acid and yeast extract elicitation on low – molecular antioxidants andantioxidant activity of marjoram (*Origanum marjoram* L).Acta Scientiarum Polonorum – Technologia Alimetaria; 2017.16(4)371-377.

90-Preeti Sharma; Pradeep Kumar; Rachna Sharma; Gaurav Gupta; Anuj Chaudhare. Immunomodulators: role medicinal plants in immune system. Nationl journal of physiology, pharmacy and pharmacology; 2017. 7(6): 552-556.

91-Ferreira, T.P.deS.;Veloso, R. A.; Santos, G. R. dos; Santos, L.P. dos; Ferriet, T.P. de S.; Barros, A. M.; Possl, R.D.; Aguiar,R.W. de S. Enzymatic activity and elicitor of phytoalexins of lippie sidoides Cham and endophytic fungi.African journal of Biotechnology 2018.17(15):521-530.

92-Kiran Sharma, Rasheeduz Zafar .(2016) . Optimization of methyl jasmonate and-cyclodextrin for enhanced duction of taraxerol and taraxasterol in (*Taraxacum officinale* Weber) cultures Plant Physiology and Biochemistry 103 ,24e30 .

93-Pavela, R., 2011. Insecticidal and repellent activity of selected essential oils against of the pollen beetle. Meligethes Aeneus 34, 888–892.

94-Pavela, R., Vrchotova, N., Triska, J., 2009. Mosquitocidal activities of thyme oils (Thymus vulgaris L.) against Culex quinquefasciatus (Diptera: culicidae). Parasitol. Res. 1051365–1370.

****