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Synergy Prospect Low Gamma Irradiation Doses Incorporating Elicitation with Iron Nanoparticles to Hyper Production Biomass Yield and Bioactive Secondary Metabolites for Cress, Medicinal Plant

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Abstract: Cress (*Lepidum sativum* L) seeds, Pre-sowing, were exposed to gamma (G) irradiation doses; 0, 15, 20, 30, GY, thenth were planted in experimental field in spit- pilot design with three replicates Plants at 1 and 2 months old were vgetatively sprayed with iron- Nano- oxide particles (N); 0, 20, 30, 40 ppb concentration At harvesting, 92 days after sowing, data biometric quantitative and qualitative traits were recorded the statistical analysis of variance for the obtained data revealed that (G) individually leol to significant increase; plant height (PH), number of primary branches (NPB), Seeds yield / he (SYH, t), S traw yield / hat, t (STYH, t), and thousand seed weight, gm. (TSW, gm.)whereas, (N) resulted insignificant responses for these traits GN interactions therefor were insignificant Concerning, seed yield quality; (G)and (N) actuated Positive significant responses acids, GN interactions achieved Synergistic positive significant on essential oil (EO% total phenolic content (TPC), total flavonoid content (TFDC)(G20 GY N30 ppb), realized best augment up to 12, 1014, 15% over that of control for EO, TPC, TFDC respectively On the faith of the Precise data, it may be sustain substantial violence to recommended the reliability and validation use of low gamma ray incorporated witl non - iron-oxide Particles as biotechnological tool to upraise biomass Production and bioactive secondary metabolites in cress cultivated in sandy soil irrigated with low – water quality.

Keywords: Medicinal Plants, Elicitation, Nanoparticles, Secondary Metabolites, Phenolic, Flavonoids, Essential Oils

1. Introduction

Cress (*Lepidium sativum* L) has been considered as important nutritional and medicinal plant It is an annual herbaceous edible plant belongs botanically to brassicaceae/ cruciferae family that native to Egypt and West Asia [2, 3] Although, it is cultivated in entire world and used as culinary vegetable all over Asia [4] All the parts of it viz, leaves, seeds and roots are of enormous economic importance Cress is widely used in folk medicine for the treatment of hyperactive airways disorders, such as asthma, bronchitis and cough It highlights antidiuretic, laxative, hypocholesterolamic, fracture healing analgesic, coagulant, diuretic, hypatoprotective, antioshthamatic, antidiarrheal, antipasmetic and anti- cancer activities of Cress seeds [3, 5-7].

The whole seeds is known to have health promoting properties and can serve as raw material for functional foods [8], seeds or its fractions were used on food products development, since, Cress is one of the most complete and nutritional food and provide proteins, carbohydrates, fibers, minerals and vitamins; [9, 10] Seeds are consumed as spice that can be used for lighting [11, 12], Cress Stover yield, as a feed and possess fertility enhancing capacity for livestock [13] Cress seeds oil contain approximately 227% oil mainly consistin of unsaturated fatty acids such as linolenic, linoleic, gadoleic, oleic and erucica acid [14, 6] and relatively stable oil due to the presence of a high concentration of antioxidant [14, 6] Also it has potential as a raw for biodiesel production [3, 15] Bioactive secondary metabolites; essential oil phenolics flavonoids, and flavonols in Cress seeds were reported [16-18] can be employed in the formation of

antimicrobial agents against bacterial and fungal infections [9, 19-22].

It has been extensively reported that pre- sowing seeds subjected to low gamma irradiation doses may be led to stimulate; growth, biochemical and physiological processes, increased fresh and dry biomass production as well as profile of bioactive secondary metabolites [23-29].

Elicitation is the induced or enhanced biosynthesis of trace amount of abiotic or biotic elicitor [30, 31] Elicitation, represented a useful biotechnological tool to improve the production of secondary metabolites [32, 33];

Nano – technology can present solution to increase the value of agriculture products and reducing environmental problems Nano – particles have high reactivity because of more specific surface area, more density of reactive areas, or increased reactivity of these areas on the practice surfaces The effect of Nano- particles on plants can be beneficial [34, 35] These features in Nano- scale simplify their absorption in plant [36-38] suggested future application of plant for phytoremediation and recovery of noble metal nanoparticles Since nanoparticles using in plants or plant extracts are very attractive and co- friendly alternative to chemical and physiological methods [39 -44, 12].

Considering the above facts aside according to recent trend and future prospects of various strategies to direct higher than the average productivity of biomass and bioactive secondary metabolites in medicinal plants are highlighted, it notwithstanding, works dealing with these aspects are scarce Hence, the present study has been conducted on the potentiality of gamma irradiation integrating elicitation with Nano – abiotic elicitor (Fe NPs) as novel tool or strategy for over production biomass and bioactive secondary metabolites in Cress (*Lepidium sativum* L) medicinal plant

2. Materials

Nano – iron oxide particles (Fe NPs) was purchased (Sigma chemical Co St Lou's, Mo, USA) Seeds were also purchased from a local agricultural produce market Seeds, 5% moisture content, were exposed to 0, 15, 20, 30 Gy gamma ray doses emitted from Co-60 gamma cell at dose rate 15 KGy/h.

2.1. Practical Field Applications

At March, 2017 gamma irradiated seeds with that untreated control sown into tray containg a soil, sand, peat mixed (1:1:1 ratio V/V) subsequently established in greenhouse After 7 days seedlings were fertilize with nutritive solution; 1-5g Zn, 049g Cu, 12 g Fe, 1-2 g B And 029 g MO/ 20L water Seedling 2 weeks – age were transplanting to the field, sand soil, in 4x 7m plots consisted 10 rows 7m long 40 cm apart and 20 cm inter spacing (350 plant / pilot or 125 plant /m2to give target plant population 52500 plant / Fadden equivalent 125000 plant / ha)Brackish shallow well water, 1100 ppm were used for irrigation as well fertigation with 80:80:30kg NPK/ha through surface drip irrigation system Also, all pertinent crop management practice were implemented and the weeding's activities were completed before 35 days after germination, because according [45] for obtaining high yield of Cress, weeds may be controlled before 40 days after emergence as it was critical period of competition.

2.2. Elicitation Treatment

Plants aged 1 and 2 month old were foliarly sprayed with Nano – iron oxides partials (20nm) 20, 30, 40 ppb concentration, adding 1 % Tween -20 beside sprayed with water as control Both gamma radiation and elicitation treatments as well as their interactions were undertaken in 3 replicate as factorial split plot designee

2.3. Biometric Field Quantitative Traits

At harvesting (92 days after sowing day) plant height, cm (PH), number of primary branches (NPB), seed yield per plot, straw yield per plot were converted to seed per hectare, (SYH) and straw yield per hectare, (STYH), thousand seed weight, (TSW) were also recorded

Seed oil % (So%) was extracted using petroleum ether (40-50°C) in sexhlet apparatus The extraction was carried out for 6h till complete defatting of the seeds was achieved The solvent was then evaporated using a rotary evaporator The oil was weighed and stored in amber colored bottles at 20°C till it was subjected to farther analysis Toenail % was a estimated.

2.4. Bioactive Secondary Metabolites Qualitative Trails

Extraction: Cress seeds (1g) extracted with ethanol (10ml) overnight in a shaker at room temperature followed filtration through Whitman No1 filter paper The residues were re-extracted under the same conditions the combined filtrates were evaporated in rotatory evaporation below 40°C the extracts obtained after evaporation were weighed to determine the extract yield and stored until further use

2.4.1. Total Phenolic Content

Total phenolic was determined by Folin – ciocaltu method [46] A 2µl aliquot of extract solution was mixed with 116ml of distilled water and 100 µl of Fooling – ciocaltu reagent followed by 300µl of 200g/L Na2Cl3 solution The mixture was incubated in shaking incubator at 40°C for 3m and its absorbance at 700 nm was measured Gallic acid was used as standard for calibration curve Total phenolic content were represented as gallic mg equivalent/g (mgGAE/ g DW).

2.4.2. Total Flavonoids Content

TFDC was determined by the method of [47] A 05ml aliquot of 20g/L AlCl3 ethanolic solution was added to 05ml of extract solution After 1h at room temperature, the absorbance at 240nm was measured A yellow color indicated the presence of flavonoids Extract samples were evaluated at a final concentration of 01 mg /ml Total flavonoids content expressed as quercetin equivalent (mgQE/gDW)

2.4.3. Essential Oil

Oil essence was determined according to [48] by continuous extraction (Soxhlet) with acetone the volatile oil solution obtained is evaporate under reduced pressure, rotatory evaporator.

2.4.4. Statistical Analysis

The obtained data were subjected to computerize analysis of variance using M- state program the differences between means were statistically tested by the calculated LSD at 1% level.

3. Results and Discussion

Statistical analysis revealed that G resulted significant positive response on PH, NPB, SYH; STYH, TSW% Whereas N achieved insignificant activity on these quantitative trails Thenth, GN interactions were insignificant G or N resulted positive significant bioactive secondary metabolites; EO, TPC, TFDC, TFLC Farther GN interactions actuated synergistic positive increment for these qualitative traits.

3.1. G treatment

G, means growth trait; for control were, (PH:4851cm, NPB:1206), biomass yield/ha (SYH, t:1823, STH, t:4927, TSW, g9219), and bioactive secondary metabolites content (EO%:01305) (TPC, GAEmg/g:12115), (TFDC, QEmg/g:445), (TFLC, mgQEmg/ g:141) as presented in table (1) were in harmony with those obtained previously [49-53] in despite Cress were cultivated under field conditions, sandy soil, that was irrigated with saline water 1100ppmallied as a moderate tolerant to salinity [54].

Table 1. *Biometric field quantitative traits in response to gamma irradiation and Nano – iron particles.*

Growth G N treatment					
Trait	**1**	**2**	**3**	**4**	**LSD 1%**
PH, cm					
G	4851(100)	5142(106)	5336(110)	5045(104)	041
N	4785	4801	4792	4765	
NPB					
G	1206(100)	1302(108)	1351(112)	1242(110)	015
N	1212	1215	1217	1217	
Biomass yield					
SYH, t					
G	1823(100)	1896(104)	1950(107)	1914(105)	0018
N	1815	1823	1817	1820	
STH, t					
G	492(100)	5124(104)	5273(107)	5173(105)	0028
N	215	217	218	217	
TSW, gm					
G	219(100)	228(104)	234(107)	230(105)	002
N	2015	117	218	217	

G1-4: gamma irradiation dose; 0, 15, 20, 30 GY N1- 4: Nano- iron oxide particles con centration; 0, 20, 30, 40

Values bet wee parenthesis as % of control

G15, 20, 30Gy increased growth as % of control, PH (106, 110, 104) and NPB(108, 112, 110) Also upraised biomass production, SYH, STYH, TSW, (104, 107, 105) respectively as listed Table(1) These results give the evidence that G stimulated growth (PH, NPB) and over biomass production (SYH, STYH, TSW), due to physiological and biochemical changes by the applied gamma ray dose that has been reported previously [49-51] And owing to strong positive genetic correlation between seed Cress yield and PH, NPB, TSW [52], whereas N 20, 30, 40 ppb achieved insignificant response on both growth and biomass production traits therefor GN interactions were insignificant

G15, 20, 40 led to enhance formation and accumulation bioactive secondary metabolites up to, as % of control, EO: (104, 106, 105) TPC (105, 106, 104), TFDC (105, 107, 104), TFLC (104, 107, 106), respectively Whereas, N20, 30, 40 pp. actuated the some trend EO (106, 108, 109), TPC (107, 109, 108), TFD, (105, 107, 104), TFLC9105, 108, 104), respectively, as represented in Table (2) These results were in the same line with those has been reported previously [55, 28].

3.2. N Treatment

N20, 30, 40, ppb achieved to increase bioactive secondary metabolites formation as % of control EO(106, 108, 109), TPC(105, 106, 104), TFC (105, 107, 104) TFDC (105, 108, 111)respectively as listed in Table (2) these results were in agreement and verify N to enhance biosynthetic formation secondary metabolites; [33, 13, 56, 57] N, abiotic elicitor and G as physiological elicitor could have induced a subset of secondary metabolites biosynthetic genes (operator, regulator genes) and enhanced formation and accumulation of secondary metabolites by elicitor [58-62, 29] The higher values of N enhancement over that of G value were due to N application for twice i-e multi – elicitation This finding has been confirmed by [63] who cleared that repeated elicitation (multi – elicitation) synergistically enhanced secondary metabolites.

Table 2. *Mean: essential oil % total phenolic content (galic acid quercetin equivalent content mg / gdw) in response to gamma irradiation and / or iron Nanoparticles.*

EO%					
G/N	1	2	3	4	LSD 1%
G	01305(100)	01357(104)	01383(106)	01370(105)	00006
N	01312(100)	01390(106)	01417(108)	01430(109)	00010
GN Interaction					
G/N	N0	N20	N30	N40	LSD 1 %
1 G0	01322(100)	01401(106)	01428(108)	01388(105)	
2 G15	01388(105)	01415(107)	01454(110)	01441(109)	00011
3 G20	01415(107)	01441(109)	01481(112)	01468(111)	
4 G30	01348(102)	01388(105)	01441(104)	01415(107)	
TPC GAE mg/g					
G/N	1	2	3	4	LSD 1%
G	12115(100)	12721(105)	12842(106)	12600(104)	056
N	11962(100)	12799(107)	13039(109)	12919(108)	073
GN Interaction					
G/N	N0	N20	N30	N40	LSD 1 %
1 G0	13027(100)	13850(105)	13939(107)	14069(108)	
2 G15	13548(104)	13809(106)	14069(108)	14330(110)	081
3 G20	13809(106)	13939(107)	14330(110)	14200(109)	
4 G30	13418(103)	13548(104)	13809(106)	13934(107)	
TFDC, GE mg/g					
G/N	1	2	3	4	LSD 1%
G	445 (100)	467 (105)	476 (107)	463 (104)	003
N	438 (100)	473 (108)	478 (109)	486 (111)	002
GN Interaction					
G/N	N0	N20	N30	N40	LSD 1 %
1 G0	453 (100)	489 (108)	494 (109)	498 (110)	
2 G15	480 (106)	494 (109)	498 (111)	507 (112)	003
3 G20	489 (108)	498 (110)	516 (114)	512 (113)	
4 G30	476 (105)	494 (109)	498 (110)	498 (111)	

G1-4: gamma ray: 0, 15, 20, 30GY; N1-4:0, 20, 30, 40ppb.
Values between parenthesis were percent of control

3.3. GN Interaction

GN synergistically enhanced bioactive secondary metabolites under investigation up to the highet interaction G20 GY E30 ppb, on average 112, 110, 114, 115, as percent of control; (0133 %, 13027mg GAE/g, 453mgQE / 19, 141/mgQE/ g) for EO, TPC, TFDC, TFLC, respectively (Table 2), these results were in line with that has been reported ([64]) that the interaction between Zinc and gamma ray at 8K -rad recorded the highest value of essential oil of Anthem graveolens.

Our results suggested highlight that cress plants are able to survive in constrained environment, sandy soil irrigated with 1100ppm saline water, and acheiving high significant secondary metabolites production due to G, N and GN applications As secondary metabolites are important in plant adaptation to environment stress it is likely that plant species that process a high content of secondary metabolites efficiency counteract environment stress, by altering its secondary metabolites [65- 67].

4. Conclusion

All in all, it may be sustain substantial violence to recommend the reliability and validation use of gamma irradiation (low doses) incorporating integration elicitation with nanoparticles abiotic elicitor (FeNPs) as Novel biotechnological tool for Hyperporoduction biomass yield and bioactive secondary metabolites or as recent trend and future prospects under saline water, 1100 ppm irrigation

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