

UV Radiation Duration and Biological Combination (Mycorrhiza and Azotobacter): Impacts on Jojoba Seed Germination and Growth

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Abstract. A study was conducted in agricultural season 2021 to find out the effect of UV radiation duration and Biological combination (mycorrhiza *G. mosseae* and *Azotobacter chroococcum*) (F) on the germination and growth of Jojoba (*Simmondsia chinensis*). Jojoba seeds were treated with UV for different durations of 5, 10 and 15 minutes and coded it (UV5, UV10 and UV15). Then seeds were transferred direct after being exposed to UV radiation in pots supported by Biological combination add it under seed by average 10 g every pot By "using Randomized Complete Block Design (RCBD) with three replicates". The results were analyzed statistically using SAS. The results showed significant increase in interactions UV5+F on number spores, colonization intensity and Mycorrhizal colonization percentage (37.00 g⁻¹ soil fungal spore, 55.7%, 90.00% respectively). The highest percentage of Nitrogen% and percentage of phosphorus (3.50% and 0.33%) respectively compared by control (2.33.0.21) % respectively, germination percentage (89.67%), the lowest days on germination speed (9.5 day) and high plant height (76.67 cm) compared to treatment UV15 (75% and high number days for germination (16 days) which no significant differences in the control and the last treatment low high plant (66.33 cm). The treatment UV10 +F showed a statistically significant increase on number of leaves (67 leaves) and total chlorophyll content of jojoba leaves (31.33 spad) compared to control (31 leaf, 14.33 spad). Leaf area showed no significant differences in the treatments. Results revealed that the seed treatment with biological combination and UV (particularly UV 5 improved some traits in jojoba seed.

Keywords. Gold green, Number spores of mycorrhizal, Fixing nitrogen, UV.

1. Introduction

Iraq's agricultural has suffered from a lack of water resources and a decline associated with low rainfall during the winter season in recent years, that was reflected, the lack of the increase natural vegetation where trees and shrubs that are tolerant of salinity and drought are planted in the form of green belts and windbreak [1]. "The Desertification has mainly spread in Iraq due to military operations" that have damaged soil class, that was returned to turning productive land into barren land, increased dunes, and increased pressure on groundwater to threaten water and food security [2]. So came the thought of growing plants that bear the extreme conditions of the country, such as the Jojoba

tree (*Simmondsia chinensis*), is highly valued for its drought resistance stress conditions as well as being an environmentally friendly plant for its role in the production of biofuels and medical, industrial benefits, increased shrubbery, combating desertification soil stabilization, and the possibility of using it as a green belt around cities [3]. Jojoba is one of oil crop and has been grown since ancient times in various desert regions around the world, including Egypt, Saudi Arabia, India, Chile, Australia and Mexico [4]. Jojoba plants can grow with deficit water to survive due to their ability for drought- and salinity-resistant. " Due of its purity, stability, lack of odor, and antioxidant content, jojoba oil is used as a biofuel and a natural product base for many cosmetic products[5]. Furthermore, could be utilized as a fundamental feedstock in the chemical sector such as lubricant, gear additive, medicinal, extender, anti-foaming, and wax and polish industries[6]. Furthermore, plant extracts for leaves have been displayed to elicit varying degrees of antioxidant activity imply that these extracts may serve as pharmacological agents with antioxidant properties [7]. One of the most important physical phenomena used in this area is the cheap Ultraviolet radiation and its safe impact on health and the environment, and it is an ionizing electromagnetic radiation[8]. Plant morphological and physiological responses are thought to be modulated by UV at ecologically relevant intensity [9]. Through photo destruction or enzymatic processes, it affects phytohormones and interferes with plant growth[10]. The some studies confirmed that the seeds or plant are exposed to low doses and for a few periods of harmless UV rays that stimulate and regulate many morphological, physiological and biological changes in it[8,11]. Environmentally friendly farming approaches have become more prevalent in agricultural production in recent decades. This refers to cutting back on chemical inputs without sacrificing agricultural output levels or quality. A low-cost sustainable agricultural system can be achieved by integrating soil management with biofertilizers[12]. Biofertilizers have a major role in fixing nitrogen and increasing plant nutrition by growing the surface area of the roots, which is reflected in increasing the absorption of nitrogen and phosphorus, thus achieving the nutritional balance of the plant[13]. Thus, the use of symbiotic Arbuscular Mycorrhizal and plant growth-promoting rhizobacteria is taken into consideration as a means of achieving the objectives of sustainable agriculture. Plants and AMF can successfully associate, improving both species' nutritional status and minimizing the need for fertilizers, particularly P.[14]. *Azotobacter* is a free-living in root plant that fix nitrogen and can aid in the growth of a variety of crops through the synthesis of some regular growth such as cytokinin; "gibberellic acid, and indole acetic acid (IAA) "[15]. There are two categories of PGPR mechanisms: direct and indirect. The synthesis of phytohormones (auxins, cytokines and gibberellins) the solubilization of minerals in the soil, N₂ fixation, and the reduction of ethylene levels are examples of direct methods[16]. Facilitating the colonization of other beneficial soil microbes is one example of an indirect approach. These fertilizers are produced from the isolation of microorganisms and their proliferation on appropriate farms and then carried on a suitable holder that is preserved in appropriate conditions until it is used as an inoculate for seeds, roots or soil[17]. The culture of growing jojoba plants in Iraq is weak, as are the guiding messages through research and relevant institutions. Jojoba cultivation needs a national project adopted by state institutions, with government support to benefit from Egypt's experience, which has succeeded in investing in jojoba to stimulate its green economy [18]. Therefore, "Jojoba is considered one of the practical and scientific solutions for plantation in Iraq". In spite of its importance, very few studies which interest effects of abiotic and biotic stresses on the germination seed, development and yield of the jojoba. The objective of the paper to find out the beneficial curing properties of the Biological Combination to measure seed germination and development growth jojoba as an outcome of UV Radiation pretreatment

2. Materials and Methods

The experiment was carried out in the Department of Agricultural Research-"Ministry of Science and Technology" for the agricultural season 2017. The experiment design was Randomized complete block design (RCBD) with three replicates". To study the effect UV duration and bio-synthesis in stimulating and growing seed jojoba. The biological combination(bio-synthesis) obtained from the Laboratories of the Agricultural Research-The Center for Biological and Food Technologies and was

the first inoculate of *Azotobacter chroococcum* bacteria (with a inoculate density of 1×10^9 cuf ml⁻¹) grown on Waksman no.77 medium under shake culture condition for 7 days The cultures were then centrifuged at 5000 rpm for 7 minute supernatant was discarded and the pellets containing N-fixing bacterial cells and Mycorrhizal inoculate using the reproductive parts of the fungus(spores, fine roots of maize and hypha) blends these "compositions with a peat soil was autoclaved at 0.14 MPa and 121°C for 2h". *G. mosseae* was propagated using maize as host plant inoculum was added to the seedling point at the rate of After 4 months, a mixture of spores (density > 40 spore g soil⁻¹),including infected root fragments, Mycorrhizal, were harvested. Exposed to UV rays for 0, 5, 15 and 30 minutes mediated by the UV-generating [19].The seeds were grown immediately after being exposed to UV radiation with the addition of a biological combination. The seeds were planted in pots (30× 30×2 cm) having sterilized sand: soil mixture (500:1,500 g) To each pot, Soil sample were analyzed which were estimated some chemical and physical properties Table 1.

Table 1. Some chemical and physical properties.

Subject	Value	Unit
PH	7.1	-
Ec	2.9	De Salimeter.M ⁻¹
N	8	Mg.Kg Soil ⁻¹
Total P	7.5	Mg.Kg Soil ⁻¹
K	105	Mg.Kg Soil ⁻¹
Organic Matter	6.5	G.Kg Soil ⁻¹
Sand	750	G.Kg ⁻¹
Mud	250	G.Kg ⁻¹

10 % inoculum consisting of biological combination (*G. mosseae* + *Azotobacter chroococcum*) (F) were added under the seeds planted. Each treatments had three replicates with 40 seeds in each replicate per treatment had five pot (8 seed)[20].The number of the treatments were eight treatments (35 pots),the symbol for the parameters in the study are as follows:Con(without anything),F=(Biological combination),UV 5 = Five minute for exposed seed to UV ray , UV 10= ten minute for exposed seed to UV ray and UV 15= Fifteen minute for exposed seed to UV ray. The experiments were transferred in plastic house having constant temperature (25–35°C) and Relative humidity (60–70 %) for further studies. Light was provided by sunlight under a 14-h photoperiod. Plants were watered regularly and 55 ml per pot was given every 2 day. The experiments were carried out in alone, combined inoculations. In the control, no biological combination was added and exposed to UV rays. Adopted the following gradient to measure the severity of the infection:

1=zero no infection

2=1-25% of the root total infected

3=26-50% of the root total infected

4=51-75% of the root total infected

5=76-100% of the root total infected

"Different growth and biochemical parameters were studied the severity of the injury was calculated according to the equation[21]:

$$\text{The severity of the injury} = \frac{(\text{the roots infected} \times \text{the degree of injury})}{(\text{total number of pieces examined} \times \text{the highest degree of injury})} \quad (1)$$

The percentage root colonization was calculated using the following formula [22]:

$$\text{Root colonization (\%)} = \frac{(\text{Number of arbuscularmycorrhiza - positivesegments})}{(\text{Total number of segments studied})} \times 100 \quad (2)$$

The leaves of five plants from each treatment were harvested and dried in an oven at 75°C until a constant weight. Then, the dried leaves were ground separately. Subsequently, 50 mg of each sample were employed for the measurement of N concentrations was determined by micro Kjeldhal method as outlined by[23].For the determination of the concentrations of other nutrients, including P and K. 0.2 g

of each sample was digested in 10 mL of acid mixture (HClO₄:HNO₃,1:5) and diluted with double-distilled water to 100 mL The concentration of P was determined spectrophotometric using ammonium molybdate blue method[24].Seed germination percentage was calculated using the following formula[25]:

$$\text{Germination percentage \%} = ((\text{Number of germinated seeds}) / (\text{Total number of seeds}) \times 100 \quad (3)$$

Speed of germination was calculated by the following formula.

$$\text{Germination speed} = n_1/d_7 + n_2/d_8 + n_3/d_9 \pm \quad (4)$$

Where n= number of germinated seeds, d= number of days [26].

Plant height (cm), total Leaf Number, average Leaf Area by using area meter (area meter A.M100).The chlorophyll content index was determined on the midpoint of the youngest fully-expanded leaf by using Minolta SPAD (Chlorophyll Content Meter (CCM-20) spad from company OPTI-SCIENCES The according to our previous study[27].The experimental design was complete randomized block design (CRBD) was used in this experiment with three replicates. Data obtained were analyzed using SAS Inst software. Data are shown in table 2 were analyzed with analysis of variance (ANOVA) procedure, differences between means were compared by using LSD test at $P > 0.05$. All the parameter data were analyzed with analysis of variance (ANOVA) procedure, differences between means were compared by using Duncan's Multiple Range test at $p < 0.05$.

3. Results

3.1. Parameter Mycorrhiza

Biological combination and exposed to UV ray and their interactions had significant ($p < 0.05$) effects on number spores, colonization intensity and Mycorrhizal colonization percentage (Table 2). Number spores (32 spores g soil⁻¹), colonization intensity (50%) and Mycorrhizal colonization percentage (60%) were increase when plants were inoculated with F as compared with the control. interaction between F and UV by all Durations increasing significant on Number spores, colonization intensity and Mycorrhizal colonization percentage and the best interaction treatment was five minute by UV radiation was recorded 37 spore g soil⁻¹, 55.7% and 90% respectively. (Table 2).

Table 2. The effect of Biological combination (*G. mosseae*+ *Azotobacter chroococcum*) and UV rays on the on parameter Mycorrhizal in jojoba plants.

Treatment	Number spores g soil ^{*1}	Colonization intensity %	Mycorrhizal colonization percentage%
Con	0.00	0.00	0.0
F	32.67	50.00	60.0
UV 5	0.00	0.00	0.0
UV 10	0.00	0.00	0.0
UV 15	0.33	0.00	0.0
UV 5+ F	37.00	55.70	90.00
UV 10 + F	36.00	53.33	85.7
UV 15+ F	30.33	34.00	84.2
LSD 5%	2.66	8.36	10.55

Con (without anything), F(Biological combination), UV 5 = Five minute for exposed seed to UV ray , UV 10= ten minute for exposed seed to UV ray and UV 15= Fifteen minute for exposed seed to UV ray.

3.2. Percentage of Nitrogen% and Phosphorus in Leaves

Percentage of Nitrogen% and phosphorus on leaves jojoba plants were significantly affected by interaction between Biological combination and UV radiation (Fig.1). The highest percentage of Nitrogen% and percentage of phosphorus were recorded on interaction Biological combination and

UV 5 (3.50% and 0.33%) respectively and the lowest was obtained in control (2.33% and 0.21%) respectively.

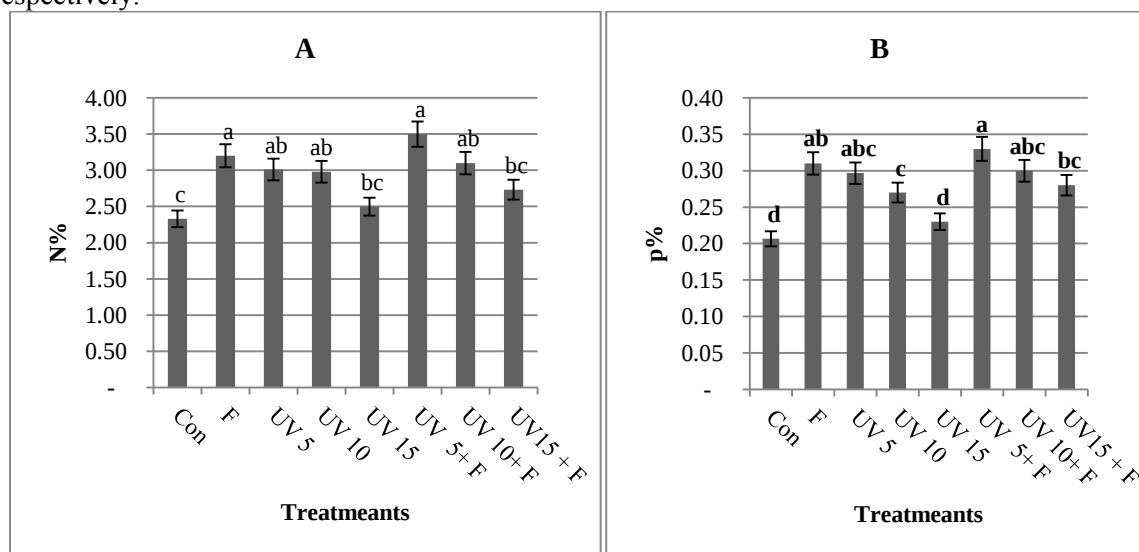


Figure 1. The effect of Biological combination (*G. mosseae* + *Azotobacter chroococcum*) and UV rays on the percentage of Nitrogen% (N) (A) and the percentage of phosphorus (P %) (B) in leaves of jojoba plants. Con (without anything), F(Biological combination), UV 5 = Five minute for exposed seed to UV ray, UV 10= ten minute for exposed seed to UV ray and UV 15= Fifteen minute for exposed seed to UV ray. Letters indicate significant according to Duncan's Multiple Range test at $p < 0.05$.

3.3. Parameter Germination

Data illustrated in Fig.2C demonstrate the effect of Biological combination and UV rays on germination rate % and germination speed (day). It is clear that germination percentage seed jojoba were increased in treatment interaction Biological combination and UV 5 rays (89.67%) the lowest that germination percentage (75%) was obtained with the treatment UV 15 and contro, No significant differences were observed between others treatments. The higher germination speed were recorded (16 day) in control and no significant differences in the treatment UV 10, The lowest value on germination speed on treatments F+ UV 5(9.5day) and no significant differences in others treatments Fig.2(D).

3.4. Parameter Growth

Growth performances of jojoba plants were estimated by growth parameters (plant height (cm), number leaves, leaf area and total chlorophyll content of jojoba leaves). The effect of Biological combination and UV rays on growth parameters of jojoba plant (Fig.3E-H) showed that these plants exhibited a significant increased ($p < 0.05$) in their growth parameters (Fig.3E), was markedly increased on plant height (76.67 cm), compared to control plants (66.33 cm). The treatment UV10+F showed a statistically significant increase ($p < 0.05$) on number leaves (67.33 leaf) and total chlorophyll content of jojoba leaves (31.33 spad), compared to control (31.67 leaf, 14.33 spad) respectively (Fig.3F and H). Leaf area was no significant differences in the treatments (Fig.3G).

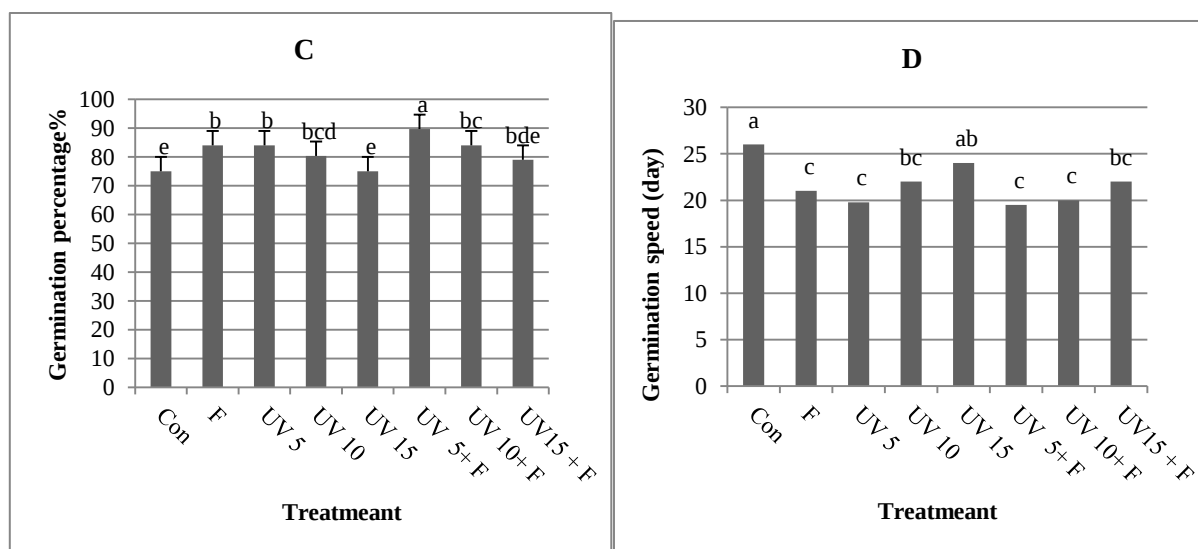
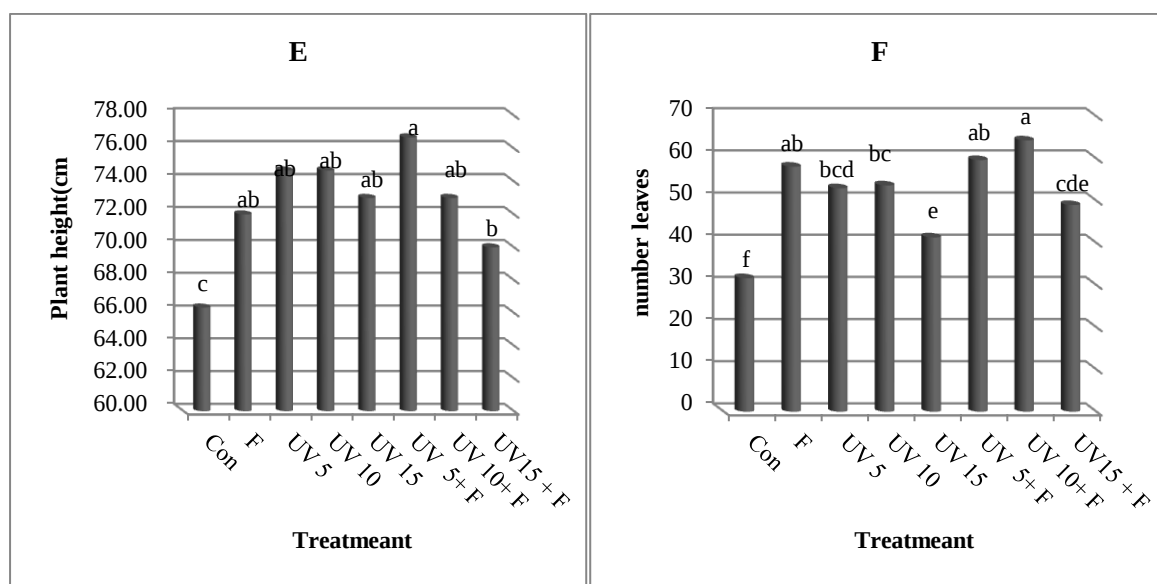


Figure 2. The effect of Biological combination (*G. mosseae* + *Azotobacter chroococcum*) (and UV rays on Germination percentage % and Germination speed (day) (D) on leaves jojoba plants. Con (without anything), F (Biological combination), UV 5 = Five minute for exposed seed to UV ray, UV 10 = ten minute for exposed seed to UV ray and UV 15 = Fifteen minute for exposed seed to UV ray. Letters indicate significant according to Duncan's Multiple Range test at $p < 0.05$.



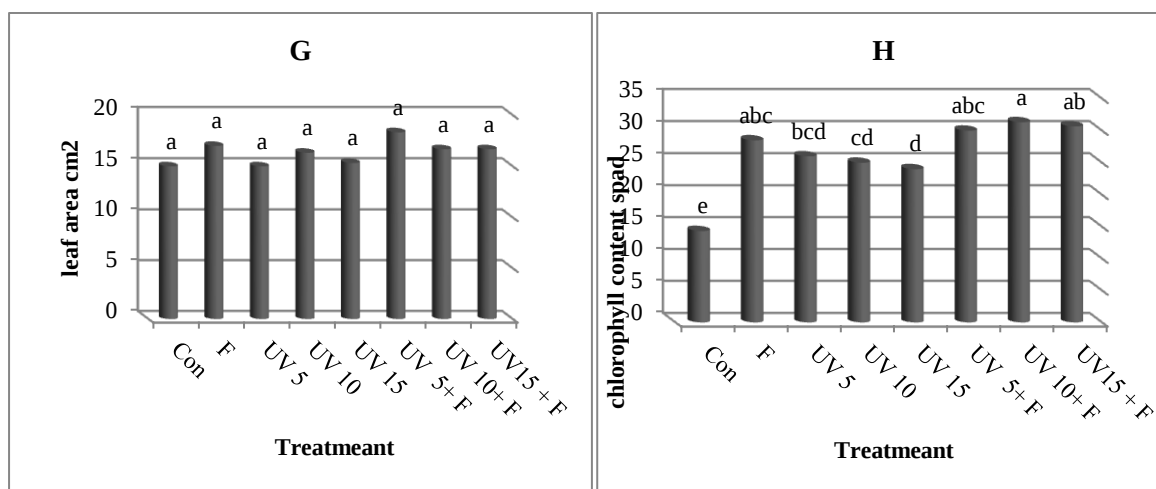


Figure 3. The effect of Biological combination (*G. mosseae* + *Azotobacter chroococcum*) and UV rays on plant high (cm)(E), number leaves (F), leaf area cm² (G) and total chlorophyll content on leaves jojoba plants spad (H). Con (without anything), F(Biological combination), UV 5 = Five minute for exposed seed to UV ray , UV 10= ten minute for exposed seed to UV ray and UV 15= Fifteen minute for exposed seed to UV ray. Letters indicate significant according to Duncan's Multiple Range test at $p < 0.05$.

4. Discussion

In general, the results of the research showed a relationship between the jojoba plant and biological combination (F) after 24 months from inoculation, especially plants treated with UV radiation, Because the Simmondsiaceae family have a familiarity with Mycorrhizal fungi and *Azotobacter chroococcum*, and that plants were treated with UV rays are more likely to be infected than others. The treatment F+ UV5 had increased spore numbers g.soil⁻¹, colonization intensity %, and Mycorrhizal colonization percentage% (Table2). There was a correlation in the number of spores in the root zone soil (32spoers g soil⁻¹), colonization intensity (50%), and Mycorrhizal colonization percentage (60%) when plants were inoculated with F(*G.mossa*+*A. chroococcum*) as compared with the control, supporting the by[28].The authors implie a synergistic activity in the biological combination (*G. mosseae* + *Azotobacter chroococcum*), which increases the activity of *G. mosseae* by generating organic acids, which act as a carbon source for the fungus, or by generating by synthesizing antioxidant enzymes including, peroxidase, catalase, superoxide and non-enzymatic antioxidants such as glutathione, ascorbate and α -tocopherol; hence, providing an suitable way to replace the hazardous agricultural chemical and agro-ecosystems destabilizing fertilizers[29].The potential result of Mycorrhizal fungi leading to increased spore production and colonization spore number (Table2) *A. chroococcum*'s enhanced stimulate the growth of hyphae in Mycorrhizal fungi was leading to increased spore production and colonization[30] .[31]found the same results on lettuce. The effect of radiation on the activity of metabolic processes and enzymes in the germination stage and vegetable growth[32].The possible outcome of improved plant growth indicates the improvement of organisms (Table2) which had Associating beneficial soil microorganisms with plant roots may enhance plant growth and health, as indicated by[33]".The percentage of Nitrogen and Phosphorus in leaves jojoba plants was significantly affected by an interaction between Biological combination and UV radiation (Fig.1)this increase could be attributed to improved P uptake following Mycorrhizal inoculation, N-fixation, or glutamate synthesis activity by metabolism directly links nitrogen to carbon metabolism and The diverse metabolic fates of glutamate have profound effects on plant life, Only a few functions assigned to plant glutamate receptors are related to glutamate. The receptor that perceives extracellular glutamate as a danger signal is unknown[34].P is taken up by mycorrhizal hyphae and moved from roots to jojoba leaves[35].Accordingly, these results explored that the interaction between the

Biological combination and UV resulted in enhanced nutrients uptake specifically N percentage which could contribute to increasing amino acids content and thereby the protein content in jojoba leaves[36]. Changes in the germination percentage and Germination speed were found to be attributed to UV radiation treatments. The early stage of germination following seed exposing may have involved the activation of RNA or protein synthesis, which is one explanation for the stimulating effects of UV radiation on germination[37]. These results showed that increased duration UV radiation reduced beneficial effects of Biological combination in growth of inoculated plants. The results were in agreement with the previous literature that UV light for 30, 60 and 120 min decreased germination because UV suppresses the gibberellin in lettuce seeds, preventing the germination process. Because the impact of UV radiation differs depending on the stage, or it's possible that prolonged exposure to violet rays caused mutations or harm to plant cells[38]. Biological combination had positive effect on growth parameters when plants were inoculated with than control. Similar results were obtained in neem (*Azadirachta indica*) when inoculated with *G. mosseae* + *A. chroococcum* + *B. coagulans*[29]. [39]on *Ficus binnendijkii* (Amstel King) revealed that biofertilizers led to improvement in plant vigor of peach seedlings. Moreover, they added that adding nitrogen fixing bacterium led to great promotion in all plant characters under study. It was also reported that inoculation with mycorrhizal fungi significantly increased plant biomass, plant height, leaf number, leaf area of acid lime. So[40]stated that biofertilizers increased growth characteristics of flame seedless grapevines. The possible outcome of improved plant growth indicates the improvement in biomass accumulation (Fig3) and this in turn improves N %, P % uptake (Fig 2 C andD) These observations uphold the synergistic interaction between mycorrhizal fungi and *Azotobacter chroococcum*. Higher plant height (Fig3E), number of leaves (Fig 3 F) and chlorophyll content (fig 3 H) were associated with greater plant photosynthetic rates, which in turn resulted in greater biomass accumulation. Biological combination and UV had positive effect on some parameters study than control but Interaction between Biological combination and UV had higher significant effect to the most of indicator study particularly inoculated with F+UV5 and F+UV10 treatments than control. Interaction between F and UV showed that UV had the increase effect on plants were inoculated with Biological combination nevertheless interaction between F with UV had the synergistic and cooperative effect in jojoba plants in our experiment. It is widely accepted that establishment of Biological combination induces hormonal changes in the roots of the host plant [41]. Physical factors such as UV are important internal signals that adjust plant growth and development to environmental changes [19]. They also mediate plant-microbe such as plant- mycorrhizal fungi and *Azotobacter chroococcum* interactions [42]. In our study, the duration 5 and 10 minute for exposure to UV radiation increased N %, P % uptake, germination percentage, Plant height, number leaves and chlorophyll content in the plant and less days for germination speed of non-inoculated plants, but UV15 with Biological combination reduces of on germination percent.

Conclusion

The results of the research confirmed that the increase in the duration of ultraviolet radiation led to a significant increase in number spores, colonization intensity and mycorrhizal colonization percentage for jojoba plant (Table 2). This interaction between UV rays and biological composition was positively reflected in improving seed germination and germination speed. Thus, study indicators were improved growth and development. The best positive interaction was between the duration 5 and 10 minute for exposure to UV rays and biological composition increased N %, P % uptake, germination percentage, Plant height, number leaves and chlorophyll content in the plant and less days for germination speed of non- inoculated plants, but UV15 with Biological combination reduces of on germination percent. According to the results of this study that jojoba is one of the solutions to desert farming in Iraq. The results of the research are to find one of the scientific solutions with a future dimension that will contribute to increasing the yield of jojoba plants or increasing the efficiency of environmentally friendly plants tolerant of drought conditions and hard environments using biotechnology or physics. We propose to use other complementary methods beneficial curing properties of seed germination and development growth jojoba, due to their application in agriculture and increase plant production.

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Significant Statement

The Jojoba tree (*simmondsia chinemiss*), which enjoys high specifications in drought resistance, hard conditions as well as being an environmentally friendly plant for its role in the production of biofuels and medical, industrial benefits, increased vegetation, soil stabilization, combating desertification and the possibility of using it as a green belt around cities (Rad et al. 2015).

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Ethics

This article is unique and includes content that hasn't been published before. The corresponding author attests that there are no ethical concerns and that all other authors have read and approved the manuscript.

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