
The Effect of Bacillus Bacteria on Tomato Plants and Fruits

Abdramane Saleh Zene*, Tian Yongqiang and Zhang Jianqiang

School of Chemical and Biological Engineering, Lanzhou Jiaotong University, PR China.

*Corresponding author email id: Abdramanezene@hotmail.com

Abstract – The research was carried out inside a greenhouse (in cork cans). Tomato plants were grown in cork trays containing a blend of sterile agricultural soil. Virus isolation and infection of a many of tomato plants have been maintained. four strains of PGPR (kept at -85 ° C, in the refrigerator) were used against the Pathogenic in the laboratory, which are used to improve tomato plants grown under Cucumber mosaic virus (CMV) infection. Bacterial strains were applied in the treatments of (seed, seed + irrigation). Seedling were inoculated with CMV after 19 days post-transplanting. The height of plant, fresh and dry weight of foliage and roots, the number of fruits and fruit weights were determined. The results showed the treatment with bacteria promote the growth of tomato plants, and reduced the rate of stunting tomato plants.

Key words – Pseudomonas Chlororaphis MA342, Serratia Plymuthica HRO-C48, Bacillus Subtilis B2g, B. Subtilis FZB27, CMV Induced Resistance, Tomato, Promoting Growth.

I. INTRODUCTION

The bacterial species associated with plant growth is called plant growth promoting bacteria (PGPR). Plant associated microbes affect the growth, yields, of host plant and also provide physiological and environmental advantage to the plants by various means. These microbes directly or indirectly involved in growth promotion, yield enhancement, hence term used for them is plant growth promoting microorganism (PGPR) [1], [2], [3]. Studies indicated that the Plant Growth Rhizobacter Promoting (PGPR) colonizes plant roots, it improves its growth and yield directly by improving evolution. The roots thus improve the absorption of water and nutrients from before the plant, in addition to the production of plant hormones, or in an otherwise directly altering the biological balance in the root environment through stimulation for beneficial soil microbes, or suppressing bacterial and fungal diseases and nematodes present in the soil, in addition to viral diseases its use has become a common method in many regions of the world [4]. The use of PGPR to serve agriculture has become of global importance and PGPR has been studied as biological control agents and as bio-fertilizers that can be used in improving agriculture [5].

The results of one study indicated that applying aeruginosa Pseudomonas could increase soybean resistance to soybean dwarf virus and could improve and increase plant growth and yield, so it could be relied upon as one of the measures to control soybean dwarf virus on soybean plant [6], and also in the rhizosphere of the plant, root secretes an array of exudates which contain large amount of carbohydrates, lipids, and amino acids, and these actively attract microbial population and colonize the plant roots, which directly or indirectly help in growth promotion and management of diseases or stress [7]. Biological control is process in which pathogenic strain is maintained at low inoculum density either through one or more organisms' occurred naturally or by influencing the environment, host, or by introduction of one or more antagonists in mass. Generally plants exhibit two types of resistance mechanisms depending upon the external stimuli-systemic acquired resistance (SAR) and ISR. SAR is executed upon the pathogen attack over plants, while ISR is switched on through colonization by PGPR [8]. SAR is executed through salicylic acid (SA) signal and any kind of obstacle in SA accumulation may impair

SAR. On the other hand in ISR, PGPR colonization does not manifest any symptoms over host plant [9] molecular studies over arabidopsis indicated that both SAR and ISR are connected through NPR1 gene. Earlier literature suggested that in most of the cases, PGPR trigger ISR either by strengthening the structural integrity (physical and mechanical strength) of the cell wall or by altering the host physiological and biochemical reactions. This phenomenon leads to the production of defense chemicals signals such as chitinase, peroxidase, and proteinase inhibitors [10] due to the high cost and impact of fertilizers and chemical agitators the negative impacts on the environment associated with reduced plant productivity in conditions of unavailability of sources of nutrients and mineral salt necessary for plant growth and strengthening of its defense mechanisms against pathogens. It was biomaterials that stimulate growth and resistance must be studies systemic together without affecting the environment.

Therefore, this research aimed evaluation of the efficacy of four strains of PGPR in stimulating plant growth tomatoes under (CMV) infection conditions tomato plants within the protected cultivation, by taking readings on some growth parameters (plant height and lean weight and dry plant and root system), in addition to taking readings about some productivity parameters (number and weight of the fruits).

II. MATERIAL AND METHODS

2.1. Preparation of Bacterial Suspensions and Application of Bacte

The PGPR strains used in the study were grown on a tryptic soy agar (TSA) solid environment it was incubated at 28 ° C for 24 hours and then transferred to a liquid environment tryptic soy broth (TSB) (Prepared by adding 5.1 environmental powder (TSB) to 50 ml of sterile water and shaken well, then put each 20 ml of it in a beaker). The bacteria were incubated at laboratory temperature 27-30 within the liquid environment prior to a digital vibrator 180 / cycle for 24 hours. Take 2 ml of the previous culture and inoculate it with 200 ml of a liquid environment of TSB according to the above conditions. Then, the centrifugal gas was exposed at 4000 rpm and the part floating on the surface was excluded and the sediment part was preserved. The first method: Seed treatment (S) Seeds were mixed with concentrated bacterial suspension (10^{10} cfu/ml) to ensure the adhesion of bacteria to the seeds, it was placed on a digital vibrator at a speed of 180 revolutions / minute for 4 hours at a temperature of 25-30 ° C degrees, where the number of bacteria per seed ranged between (10^6 to 10^7 cfu/seed) the treated bacteria were soaked in sterile water for 4 hours and then air-dried [11]. The second method: Treatment (seed+Irrigation) after treating the seeds in the previous way, 10^{10} bacterial cells were added to each seedling of tomato plant after 10 days of transplantation.

2.2. Preparing the Viral Vaccine and Mechanical Infection

Prepare the viral vaccine by crushing infected tomato leaves with cucumber mosaic virus, with distilled water in a ratio of (1:5) in a container of porcelain. Mechanical infection was performed in an amount of approximately 2 ml viral pollen on three real leaves of tomato plants after 19 days of transplantation, the silicon carbide powder was spread apart light on its upper leaves, and then wipe the leaves with a cotton ball wetted with the viral vaccine in one direction the leaves treated with virus vaccine were washed with the water to get rid the cell of juice residues.

2.3. Criteria for Evaluating Growth Improvement Induced by PGPR

2.3.1. Growth Criteria:

We took readings on growth parameters (plant height, fresh\ dry weight of The shoots of the plant. and the dry\fresh weight the whole of Root) at the end of the experiment. According to the following equation Stunting rate% = (healthy plant length – treated bacteria length / healthy plant length) x 100.

The rate of weight loss according to the formula next :

Weight loss rate% = (weight of a healthy plant – weight Treated bacteria / weight of a healthy plant) x 100.

According the rate of weight loss according to the formula next:

Weight loss rate% = (weight of a healthy plant – weight Treated bacteria / weight of a healthy plant) x 10.

2.3.2. Productivity Criteria

Yield parameters (number of fruits and weight of fruits) were estimated at the end experience according to the rate of decrease in the number of fruits according to the equation next:

The rate of decrease in the number of fruits% = (number of the fruits of the healthy plant - the number of fruits of the treated acteria plant / number of the fruits of the healthy plant) x 100.

According to the rate of decrease in the weight of fruits according to the equation next :

The rate of decrease in fruit weight% = (weight of fruits of a healthy plant the weight of the fruits of the treated bacteria plant / the weight of the fruits of a healthy plant) x 100.

III. RESULTS AND DISCUSSION

3.1. Plant Height

The results showed that the increase in the height of tomato plants treated with bacteria only (66.75 cm) is higher than that of the healthy plant (57.83cm) without any significant difference with non-significant superiority of B27 strain In a way (seeds + irrigation) on the rest of the strains (Table 1).

Treatment with bacteria reduced the rate of stunting of tomato plants compared to the contagious control (36.42%), the rate of stunting was the lowest (21.9%) with strain B27 by (seed + irrigation) method. The results were consistent with the results of a previous study conducted to evaluate the efficacy of the same bacterial strains included in the study in stimulating systemic resistance to a virus mosaic of cucumber in tomato plants within the protected cultivation.

The results have shown that the treatment of tomato seeds with suspensions the bacterial infection led to improved elevation of virus-infected tomato plants mosaic of cucumber where stunting rates ranged in infectious plants treatment with bacteria (5.20% -16.72%) compared to the Infectious (23.75%).

3.2. Fresh and Dry Weight of the Plants

The results indicated that the fresh weight of the shoots for tomato plants treated with bacteria only (464) g is higher than for the sound witness (317) g without any significant difference, with a non-significant superiority of the B27 strain (581.66g) by (seed + irrigation) method over the rest of the strain. It was the fresh weight of the shoots infected and treated tomatoes with bacteria (255) g are higher than in the contagious witness (131.33)

g, without significant difference, with superiority non-significant strain of B27 (seed + irrigation) method (314.66) g over the rest of the strains. The dry weight of the shoot total of tomato plants, the treatment with bacteria (5.153) g is higher than in the healthy control (33.111) g with a significant difference, with a non-significant superiority for strain B27 (33.191) g in the manner (seeds + irrigation) on the rest strains. It was the dry weight of the shoots infected and treated tomatoes (17.61) g are higher than in the contagious witness (28) g, with only significant difference with the strain B27, with non-significant superiority of strain B27 (seed + irrigation) method on the rest of the strains (Table 2).

The treatment with bacteria decreased the rate of reducing the fresh weight of the vegetative total of the virus-infected tomato plants (19.9%) compared to the infectious control (58.75%) it was the lowest rate of reduction for lean weight (0.73%) with the B27 strain in a manner (treatment Seeds + irrigation). Treatment with bacteria reduced rates of reduction the dry weight of the shoot root of tomato plants (45%) compared to the infectious control (74.84%) it was the lowest rate of reduction (35.32%), with strain B27 in the manner (seed + irrigation). Agreed the results are with the results of a previous study conducted to evaluate the efficacy of the same strains bacterial included in the study in stimulating systemic resistance to cucumber mosaic virus in tomato plants within cultivation protected. The results showed that the treatment of tomato seeds with suspensions Bacterium improved the fresh and dry weight of the population vegetative tomato plants treated with bacteria compared to control proper. It also improved the fresh and dry weight of the plants tomatoes treated with bacteria and infectious with cucumber mosaic virus compared with the infectious control, without significant differences between strains bacterial, and treatment with bacteria reduced rates of reduction in the fresh and dry weight of the shoots ranged between (32.04% - 5.07%) - (9.68% - 2.15%) respectively compared to the infectious control (39.19%) - (27.78%) respectively. A mixture of two Bacilli strains may be used as a bio stimulator to protect the plant from pathogenic and viral on the one hand and improve plant growth on the other hand [12].

3.3. Fresh and Dry Weight of the Root Assembly

The results showed that the fresh weight of the root total of plants tomatoes treated with bacteria only (55-19.33) g it may be equated with the proper witness (18.66) g. In both strains (C48.MA) it was the highest sound witness with the two strains (B2g.B27) with a significant difference with strain B27 (seed + irrigation). And it was there is a non-significant superiority of the B27 strain (55-22.33) g in a way (seeds + irrigation) on the rest of the strains. And lean weight for the root group of tomato plants infected and treated with bacteria (18-11.33) g higher than in the contagious witness (6-4.3) g without a significant difference, with non-significant superiority (18) g for strain B27 in a method (seed + irrigation) on the rest of the strains.

The results indicated the dry weight of the root total for tomato plants treated with (MA, C48) equals weight the dry weight of the healthy witness (6g), and higher than the dry weight of the witness, normal with the strains B27 and B2g without significant difference, with insignificant superiority of strain B27 (8) by (seed + irrigation) method over other subspecies. And the dry weight of the root system of tomato plants infection and treatment with bacteria (5.4) g were higher than in control infectious (2.6) g, with an insignificant superiority (5.8) g for the B27 strain by applying (seeds + irrigation) to the rest of the strains. (Table 3) treatment with bacteria decreased the rate of reducing the fresh weight of the root system of tomato plants (21.4%) Compared to the infectious control (76.95%) the lowest rate of reduction was with strain B27 in the manner (seed + irrigation). (3.53%) treatment with bacteria reduced the rate of dry weight reduction of root population for tomato plants co-

-mpared to the contagious witness (5.66%) the lowest reduction was with the B27 strain (0%).

The results are consistent with results of a previous study conducted to evaluate the efficacy of the same bacterial strains included in the study in stimulation, systemic resistance to cucumber mosaic virus in tomato plants within the protected agriculture. The results showed that the treatment of seeds tomatoes with the bacterial strains included in the study led to improves the fresh and dry weight of the root system of tomato plants. The four non-infectious and treated bacterial strains, also resulted to improve the fresh and dry weight of the root system of your plants infected and treated tomatoes without significant difference between strains.

Treatment with bacteria reduced the rates of weight loss soft and dry of the root group, as they ranged between (12.16%-8.55%) – (13.69%4.10%) respectively compared to the infectious control (17.80%) – (59.33%) respectively the results were consistent with what he indicated [12]. The colonization of plant roots by surrounding bacteria Specific rhizome improves plant growth and stimulates prolonged systemic resistance Abroad spectrum of plant pathogens, with an acid role Jasmonic rootocean containing *Pseudomonas* 06 chlororaphis is to stimulate plant growth and induce resistance Mosaic against the discovered cucumber in tobacco. *Bacillu subtilis* 21-1 BS21-N-To1 [13]. It can be effective as one of the biological control agents to curb disease in four crops by stimulating systemic resistance and improve plant growth. Dipping the seeds of peas resulted in a suspended component from the leguminosarum Bacteria. *Rhizobium* and *P fluorescens* and their mixtures to improve growth parameters (lengths Plants, dry weight, and dry weight of shoots and the root system) in plants obtained from these seeds.

4. *The Number and Weight of Fruits*

The results showed that the number of fruits of tomato plants treated with bacteria only (11.66) the fruit is higher than him in the healthy plant (8.66) a fruit without a significant difference, with an insignificant superiority of the B27 strain by (seeds + irrigation) (14) fruit over the rest of the strains (7.33) a higher fruit than in the contagious plant (5) is a fruit without significant difference, with non-significant superiority of strain B27 method (seeds + irrigation) (8.33) fruit on the rest of the strains. The weight of the fruits of tomato plants treated with bacteria only (154.3) g on it in the proper plant (105.33) g the superiority of the B27 strain in two ways (seeds + irrigation) (197.33) g respectively, in a significant way over the rest of the subspecies. The fruit weight of the infected tomato plants and bacterial treatment (85.7) g respectively higher than in the contagious witness (54.66) g and the B27 strain outperformed (seed + irrigation) (197.33) g not significant to the rest of the strains, without a significant difference between the strains (Table 4). Treatment with bacteria decreased the rates of reduction in the number of tomatoes (15.36%) compared to the infectious control (42.26%) and the lowest rate of reduction in the number of fruits was with the B27 strain, using (seeds + irrigation) (5.7%). The treatment with bacteria reduced the rate of weight reduction of tomato fruits compared to the infectious control (48.10%) the lowest rate of reduction was with the B27 strain (Seeds + irrigation) [11].

Tomato plants treated with three strains of PGPR significantly reduced the incidence of infection with cucumber mosaic virus and significantly improved yield compared to infectious and untreated plants. Another study indicated that tomato plants were tested with each of the biological preparations containing the bacteria *B pumilus* E34, *B. amyloliquefaciens* IN937a, *B. subtilis* IN937b, *B. pumilus* INR7, *B. pumilus* T4, where the shape and development of the control plants was the largest with 10 plants of the experiment, and upon exposure to (CMV), all plants in the treatment of biological preparations and treatment of the largest control with 10 days

were clearly better in terms of height, dry weight and number of flowering and the fruits are higher than those plants infected with (CMV) of the same age, which indicates that treating tomatoes with biological preparations leads to a significant improvement in growth and in the prevention of (CMV) infection [14].

Table 1. The effect of the four bacterial strains on plant height and stunting rate in tomato plants infected with cucumber mosaic virus.

Experiment	% Stunting Rate	Plants Height / cm
Healthy plant		57.83
CMV	42.36	33.33
B27 (S)		70.19
B27 (S+I)		72.25
B27+ CMV (S)	11.37	51.25
B27+CMV (S+I)	(9.21)	52.5
B2g (S)		61.66
B2g (S+I)		62.75
B2g+CMV (S)	12.67	50.5
B2g+CMV (S+I)	12.39	50.66
MA (S)		63.08
MA (S+I)		63.5
MA+CMV (S)	15.85	48.66
MA+CMV (S+I)	15.70	48.75
C48 (S)	1	61.25
C48 (S+I)		61.58
C48+CMV (S)	16.99	48
C48+CMV (S+I)	50.58	50.58

MA: *Pseudomonas chlororaphis* MA342, B27: *B. Subtilis* FZB27, C48: *Serratia. plymuthica* HRO-C48 ,B2g: *Bacillus subtilis* B2g. CMV: Mosaic virus cucumber S: seeds, S+I: seeds + irrigation.

Table 2. The effect of the four bacterial strains on the fresh and dry weight of the shoots and the rate of their reduction in tomato plants infected with cucumber mosaic virus.

Experiment	Fresh Weight of the Plants(g)	Dry Weight of the Plants	Reduction Rate in Fresh Weight of Plants%	Reduction Rate in Dry Weight of Plants%
Healthy plant	317	111.33		
CMV	131.33	28	58.57	74.84
B27 (s)	506	183		
B27 (s+i)	581.66	191.33		
B27+ CMV (s)	257.33	70	18.82	37.12
B27+CMV (s+i)	314.66	72	0.73	35.32
B2g (s)	406.66	143.66		

Experiment	Fresh Weight of the Plants\g	Dry Weight of the Plants	Reduction Rate in Fresh Weight of Plants%	Reduction Rate in Dry Weight of Plants%
B2g (s+i)	437	151		
B2g+CMV (s)	222.66	58	29.76	47.90
B2g+CMV (s+i)	300.66	62.66	5.15	43.71
MA (s)	346.33	115.66		
MA (s+i)	352	159		
MA+CMV(s)	193.33	50.33	39.01	54.79
MA+CMV (s+i)	224.33	51.33	39.23	53.89
C48 (s)	376.66	142.6		
C48 (s+i)	437	171.33		
C48+CMV (s)	195.66	50.66	38.27	54.49
C48+CMV (s+i)	260.33	56	34.91	49.69

MA: Pseudomonas chlororaphis MA342, B27: B. subtilis FZB27, C48: Serratia.plymuthica HRO-C48, B2g: Bacillus subtilis B2g. CMV: Mosaic virus cucumber S: seeds, S+I: seeds + irrigation.

Table 3. The effect of the four bacterial strains on the fresh and dry weight of the root total and the rate of their reduction in tomato plants infected with cucumber mosaic virus.

Experiment	Fresh Weight of the Plants\g	Dry Weight of the Plants\g	Reduction Rate in Fresh Weight of Plants%	Reduction Rate in Dry Weight of Plants%
Healthy plant	18.66	6		
CMV	4.3	2.6	76.95	56.66
B27 (s)	22.33	7.33		
B27 (s+i)	55	8.66		
B27+ CMV (s)	15.33	5.66	17.84	5.66
B27+CMV (s+i)	18	6	3.53	0
B2g (s)	20	6.33		
B2g (s+i)	23.66	6.66		
B2g+CMV (s)	12.66	5.66	32.15	5.66
B2g+CMV (s+i)	15	5.66	19.61	5.66
MA (s)	21.16	6		
MA (s+i)	21.66	6.5		
MA+CMV (s)	11.33	3	39.28	50
MA+CMV (s+i)	14..33	4.66	23.20	22.33
C48 (s)	19.33	6		
C48 (s+i)	22.66	6.66		

Experiment	Fresh Weight of the Plants\g	Dry Weight of the Plants\g	Reduction Rate in Fresh Weight of Plants%	Reduction Rate in Dry Weight of Plants%
C48+CMV(s+i)	14	4.33	24.97	27.83
C48+CMV(s+i)	14.66	5.66	21.43	5.66

MA: *Pseudomonas chlororaphis* MA342, B27: *B. subtilis* FZB27, C48: *Serratia plymuthica* HRO-C48, B2g: *Bacillus subtilis* B2g. CMV: Mosaic virus cucumber S: seeds, S+I: seeds + irrigation.

Table 4. Effect of the four bacterial strains on productivity of tomato plants infected with cucumber mosaic virus.

Experiment	Number of Fruits / Fruit	Fruit Weight / g	The Rate of Reducing the %Number of the Fruits	The Rate of Reducing the Weight of the Fruits
Healthy plant	8.66	105.33		
CMV	5	54.66	42.26	48.10
B27 (s)	13	168.66		
B27 (s+i)	14	197.33		
B27+ CMV (s)	8	94.5	7.6	10.28
B27+CMV (s+i)	8.33	100	3.81	5.03
B2g (s)	9.66	111.66		
B2g (s+i)	12.33	139		
B2g+CMV (s)	6.5	83	24.94	21.20
B2g+CMV (s+i)	6.66	84.33	23.09	19.93
MA (s)	9.33	111.33		
MA (s+i)	10.66	161.33		
MA+CMV (s)	7.33	91.33	15.35	13.29
MA+CMV (s+i)	8	91.66	7.62	12.97
C48 (s)	10	131.33		
C48 (s+i)	12.33	165		
C48+CMV(s+i)	6.33	71.33	26.90	32.27
C48+CMV(s+i)	7.66	81	11.54	32.09

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IV. CONCLUSION

In general, *Bacillus subtilis* FZB27 is effective and adequate in ability to improve the growth and productivity standards of tomato plants under conditions infection with cucumber mosaic virus. Through previous experiments, we found that the *Bacillus subtilis* FZB27 is superior to the rest of the strains in the

treatment process, and we obtained satisfactory results for the research , and we summarize the results in six points:

1. Treatment with bacteria reduced the rate of stunting of tomato plants treated with bacteria and infectious with the virus compared to the contagious only, and the lowest rate of stunting was with the strain, *Bacillus subtilis* FZB27.
2. Treatment with bacteria reduced the rates of reducing the fresh and dry weight of tomato plants compared to the infectious, and the lowest rate of reduction was with the strain, *Bacillus subtilis* FZB27.
3. Treatment with bacteria reduced the rate of reducing the fresh and dry weight of the root system of tomato plants compared to the infectious, and the lowest rate of reduction was with baskets, *Bacillus subtilis* FZB27.
4. Treatment with bacteria decreased the rate of reduction in the number of fruits tomatoes compared to the infectious, and the lowest rate of reduction was with *Bacillus subtilis* FZB27.
5. Treatment with bacteria reduced the rate of weight reduction of fruits tomatoes compared to the infectious, and the lowest rate of reduction was with the strain *Bacillus subtilis* FZB27.
6. Strain B27 was the best strain to use, followed by B2g, C48 and MA in the ability to improve growth parameters and productivity of tomato plants under the conditions of cucumber mosaic virus infection.

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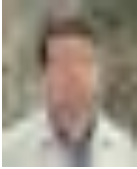
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AUTHOR'S PROFILE



First Author

Abdramane Saleh Zene, (1990), male, native place: chad (NDjamena). Candidate, Research field bio control of pathogenic bacteria. A student at a Lanzhou jiaotong university in Lanzhou\ Gansu, China.
email id: Abdramanezene@hotmail.com



Second Author

Tian Yongqiang, Male, (1972), Native place Lanzhou, Gansu PhD, Professor, Research field antagonistic microorganism and agricultural biological drugs, China. email id: tian 2918@163.com



Third Author

ZHANG jianqiang, Male, (1991), Native place: Dingxi Gansu, M.S. Candidate research field bio control of pathogenic bacteria. A student at a Lanzhou jiaotong university in Lanzhou\ Gansu, China. email id: 1556199273@qq.com