

## EFFECT OF GLUCOSE CONCENTRATION AND TEMPERATURE ON EFFICACY OF INOCULATED BACTERIA IN IMPROVING MAIZE (*Zea mays*).

*Efeito da concentração de glicose e da temperatura sobre a eficácia da inoculação de bactérias no melhoramento do milho (Zea mays)*

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### **Abstract**

Effect of bacterial inoculation of four strain of *Escherichia* (E1, E2, E3, E4) and five strains of *Bacilli* (B1, B2, B3 B4, B5) on growth of maize (*Zea mays*) var EV-6098 was studied at three glucose regimes, viz 0, 1.25, 2.50 mg g<sup>-1</sup> and at three temperatures viz. 25°C, 37°C and 39°C. Shoot growth in terms of shoot length and number of leaves per plant was better at 25°C than at 37°C and 39°C. At 0 mg g<sup>-1</sup> glucose concentration E3, B2 and B3 inoculation while at 2.50 mg g<sup>-1</sup> concentration E3 and B4 inoculations significantly enhanced shoot length, number of leaves per plant was increased significantly by B4 inoculation at 37°C and 0 mg g<sup>-1</sup> glucose concentration. All the inoculated strains of *Escherichia* at 0 mg g<sup>-1</sup> and B<sub>3</sub> strain of *Bacillus sp* at 2.50 mg g<sup>-1</sup> glucose level and at 25°C significantly enhanced root length. At 0 mg g<sup>-1</sup> of glucose concentration E<sub>1</sub>, E3 caused positive effect on seedling dry biomass at 39°C.

**Keywords:** Bacteria; Glucose; Temperature; Maize growth.

### **Resumo**

O efeito da inoculação bacteriana de quatro cepas de *Escherichia* (E1, E2, E3, E4) e cinco cepas de *Bacilli* (B1, B2, B3 B4, B5) no crescimento de milho (*Zea mays*) EV-6098 foi estudado em três concentrações de glicose, 0, 1,25 e 2,50 mg g<sup>-1</sup> e sob três temperaturas 25°C, 37°C e 39°C. O crescimento foi avaliado em relação ao comprimento e ao tamanho das folhas por planta, sendo melhor a 25°C. Sem glicose os inóculos E3, B2 e B3, e na concentração de 2,50 mg g<sup>-1</sup> de glicose os inóculos E3 e B4 aumentaram significativamente o comprimento da planta. O número de folhas por planta foi aumentado significativamente pela inoculação da cepa B4 a uma temperatura de 37°C, sem glicose. Todas as cepas inoculadas com *Escherichia*, sem glicose, e a cepa B3 de *Bacillus sp* a uma concentração de 2,50 mg g<sup>-1</sup> de glicose, na temperatura de 25°C, aumentaram significativamente o comprimento da raiz. E1 e E3, sem glicose, causaram efeito positivo na secagem de biomassa de sementes a 39°C.

**Palavras-chave:** Bactéria; Glicose; Temperatura; Crescimento de milho.

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## Introduction

The gram-negative bacterial species *Escherichia* are members of the family *Enterobacteriaceae* mostly involved in biofilm formation (1). *Bacilli* are spore-forming Gram positive rod-shaped bacteria. They are highly tolerant of adverse ecological conditions. *Bacillus* species comprise one of the most common soil bacteria groups and they are frequently isolated from the rhizospheres of plants. *Bacillus* species are also common plant endophytes (2, 3, 4, 5, 6). Because of their spore-forming ability, plant growth promoting *Bacillus* strains are readily adaptable to commercial formulation and field application (7).

Different carbon sources play major role in the growth of various plant organs. Carbon sources may result in cellular and DNA degradation resulting in increased variation in the ploidy. (8). Sugars control the expression of many plant genes and their connection to metabolic and developmental processes is unequivocal (9). Metabolic characteristics of the microbial community and the cellular composition (wall components, extracellular components) may have the potential to determine the rate and dominance of different carbon stabilization pathways. Soil microbial communities vary in their carbon processing characteristics (10). So in assessing microbial functional and taxonomic diversity, investigating community control over carbon partitioning, and quantifying the contribution of microbial cell walls to stable carbon. An important component of the rhizosphere is the actively growing microbial population, which thrives due to the provision of organic nutrients in root exudates. In turn, the microorganisms that colonize the rhizosphere profoundly affect root and plant biology in relation to nutrition, development and health. Current knowledge of root development and physiology, however, essentially comes from studies that did not take into account the effects of the rhizosphere (11).

On the other hand, the influence of the rhizosphere micro flora on root biology is so important that it is crucial to evaluate how the basic nutritional and developmental processes of the plant are affected. Soil temperature effect on all of the physical, Chemical and biological events in soils. Biological processes as the uptake of the nutrients and water by roots. The decomposition of organic matter by soil microorganism, germination of seeds

and physical processes, such as water content and movement are affected by soil temperature. Soil water content is the other important factor for plants and vegetation. For this reason, it is important to know why provide optimum temperature conditions to protect readily available water in soil. (12).

Environmental factors greatly influence the plant growth and microbial activity. The temperature range that supports plant growth is generally from 40-97 degrees F. Optimum temperatures for growth vary with the species and the stage of development and usually fluctuates night to day (13). The present research work was designed to observe the effect of temperature on growth of 10 days old maize (*Zea mays*) seedlings. The effects of temperature (25°C, 37°C, 39°C) on plant growth in combination with glucose concentration and bacterial inoculum.

## Materials and methods

### Experimental setup

Nine bacterial strains, E<sub>1</sub>, E<sub>2</sub>, E<sub>3</sub>, E<sub>4</sub>, B<sub>1</sub>, B<sub>2</sub>, B<sub>3</sub>, B<sub>4</sub> and B<sub>5</sub> were used in these experiments, which were isolated from different sources. Bacterial cultures were grown on L-Agar at 37°C for 24 hours. Effect of bacterial strains i.e. *Escherichia* and *Bacillus* isolated from biofilms and soil respectively in combination with carbon source different temperature (25, 37 and 39°C) were studied. Seeds of maize var EV-6098 were achieved from National Agricultural Research Centre Islamabad.

### Processing of seed

Healthy seeds of maize var EV-6098 were surface sterilized by soaking the seed in 0.1% HgCl<sub>2</sub> solution of five minutes. Then seeds were washed repeatedly (5-6) times with sterilized distilled water to remove traces of HgCl<sub>2</sub>. Seeds were further soaked in distilled water for 20 minutes before keeping them for germination. Petriplates (120mm) were washed and dried. Each petriplate was lined with two layers of whatman filter paper no 1. Plates were autoclaved and oven dried. 10 ml of sterilized distilled water was poured in each petriplate under sterilized conditions. 20 seeds were uniformly placed aseptically in each petriplate. Then plates were placed in dark at 25±1°C for germination. 96 pots

were washed and dried at room temperature, and then 120gm of sieved (sieve pore size) agricultural soil was taken in each pot. Carbon source i.e., glucose was mixed in soil at concentrations of 1.25 and 2.50 mg gm<sup>-1</sup> of soil.

### ***Inoculum preparation***

All bacterial cultures were grown initially for 24 hours at 37°C. After 24 hours cells were harvested. Optical density of all bacterial suspension was adjusted to 0.5 at 600 nm. Twenty-five germinated seeds were soaked in each bacterial suspension for 20 minutes. After inoculation, the inoculated seeds were transplanted in the already filled pots (having soil and different concentration of carbon source i.e. glucose). Then the effect of varying temperature (25, 37 and 39°C) was checked. For studying the effect of temperature, pots were placed at 37°C, 25°C and in wire house under natural conditions (approximately temperature was 39°C).

### ***Experimental design***

There were total 90 treatments, i.e. 9 bacterial treatments, one control treatment, three concentration of carbon source (0, 1.25, 2.50 mg gm<sup>-1</sup>) of soil of glucose and three temperatures. Experiments were performed in triplicates. Pots at 37°C and 25°C were kept at 10k lux light at 25±1°C while for 10 days. Pots were watered with distilled water. All the pots were completely randomized design at 10k lux light at 25±1°C temperature for 10 days. Experiments were performed in triplicates.

After 10 days harvest was taken. Seedlings were removed care fully from pots on eleventh day of shifting to light. Following parameters were studied. (i) Shoot Length (cm). (ii) Number of Leaves (iii) Root Length (cm). (iv) Number of Roots. (v) Dry biomass of seedling (gms).

## ***Results***

### ***Effect of inoculation on shoot growth***

All of the temperature and glucose concentration affect the efficacy of inoculated

bacterial strains in improving shoot length of maize (Table 1). Among two growth temperatures 25°C was found better than 37 and 39°C, all glucose concentrations, both for control and inoculated treatments. At glucose concentration 0 mg g<sup>-1</sup>, E3, B2 and B3 inoculation while at 2.5 mg g<sup>-1</sup> glucose concentration E3, B4 inoculation enhanced shoot length significantly over corresponding control treatments at 25°C. Conversely at glucose concentration 1.25 mg g<sup>-1</sup> none of the bacterial inoculation exhibited significant increase in shoot length over control. Maximum shoot length was achieved in E3 inoculated seedlings at glucose concentration of 2.50 mg g<sup>-1</sup> followed by B3 inoculation at 0 mg g<sup>-1</sup> glucose level (Table 1).

Generally number of leaves per plant was greater at 25°C than 37 and 39°C. However difference was not as much pronounced as in shoot length. Effect of inoculation on number leaves at 25°C was insignificant at all the three glucose regimes. At 37°C B4 inoculation significantly enhanced number of leaves at 0 mg g<sup>-1</sup> glucose level.

### ***Effect of bacterial inoculation on root growth***

In general, not always, root length at 25°C was markedly greater than at 37 and 39°C. At 25°C all the inoculated strains of *Escherichia* significantly enhanced root length at glucose concentration while inoculation at other glucose level either suppressed root length or exhibited an insignificant effect. Among the *Bacillus* sp strains, only B3 inoculation significantly increase root length at 25°C glucose level (Table 3). Generally there was not any pronounced difference in number of root between 25°C, 37°C and 39°C. At 25, inoculation of *Escherichia* and *Bacillus* sp strain either significantly reduce number of roots or exhibited insignificant effect at 0 mg g<sup>-1</sup> and 1.25 mg g<sup>-1</sup> glucose level. However, at 2.5 mg g<sup>-1</sup> glucose level all the strain of *Bacillus* sp, and E2 and E4 strain of *Escherichia* significantly enhanced number of roots. Highest response was observed due the B2 followed by B1 inoculation (Table 4). At 37°C, only E1, E2 and B5 inoculation exhibited significant positive effect on the studied parameter at 1.25 mg g<sup>-1</sup> glucose level (Table 4).

Bacteria play a major role in soil quality

**TABLE 1 - Effect of bacterial inoculation on shoot length of maize var. 6098 at different temperatures (°C) and concentrations of glucose (mg g<sup>-1</sup>)**

Treat.	25°C			37°C			39°C		
	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>
Cont	14 l-n	19.8 b-c	16 g-k	8.9b	7.6b	7.0b	7.5c-g	8.4a-d	9.8a
E <sub>1</sub>	11.3 o-p	16.4 g-k	16.5 e-j	9.0 b	10.3b	8.8b	5.2j-k	5.5h-k	5.8h-k
E <sub>2</sub>	17.3 d-i	17.4 d-i	13 m-o	6.7b	11.6b	8.2b	6.0g-k	6.0f-i	5.2f-j
E <sub>3</sub>	19.3 c-d	16.5 f-k	23.2 a	8.1b	7.8b	7.5b	7.9c-f	7.2d-h	6.8f-i
E <sub>4</sub>	10.6 p	17.8 c-h	16.1 g-l	5.8b	6.8b	12b	4.7k	7.4c-g	6.9d-h
B <sub>1</sub>	14.9 j-m	19.0 c-d	15.7 h-l	4.3b	8.9b	6.9b	6.0g-k	6.8f-i	8.3b-e
B <sub>2</sub>	18.9 c-e	17.3 d-i	16.1 g-l	9.3b	8.7b	7.9b	6.6f-j	9.7ab	6.0g-k
B <sub>3</sub>	21.9 b	18.1 c-g	14.4 k-m	6.3b	9.0b	9.0b	5.3 i-k	4.9k	5.2k
B <sub>4</sub>	15.2 i-l	18.8 c-f	19.7 b-c	8.0b	10.1b	11.3b	8.7a-c	7.7c-f	6.0h-k
B <sub>5</sub>	11.8 n-p	21.6 b-c	18.6 g-k	5.7b	6.4b	7.1b	6.9e-h	6.1g-k	6.5f-j

For each temperature value with different letters show significant difference s determined by DMR Test.

**TABLE 2 - Effect of bacterial inoculation on number Of leaves of maize var. 6098 at different temperatures (°C) and concentrations of glucose (mg g<sup>-1</sup>)**

Treat.	25°C			37°C			39°C		
	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>
Cont	2.9a-d	3.2a	3.0a-c	2.1c-h	1.3i	1.6g-i	2.0a-c	1.5c-g	1.4c-g
E <sub>1</sub>	2.8a-f	2.8a-f	2.3h	2.4a-f	2.5a-e	2.0d-i	1.9a-d	1.5c-g	1.2c-g
E <sub>2</sub>	3a-c	2.7b-f	3.0a-c	2.6a-d	2.9ab	1.4hi	1.7b-f	1.4c-g	1.3d-g
E <sub>3</sub>	2.4f-h	2.9a-d	2.6d-h	2.2b-g	1.8e-i	2.0f-i	1.1fg	1.3d-g	1.3d-g
E <sub>4</sub>	2.9a-d	3.0a-c	2.8a-f	1.5g-i	1.8e-i	2.1c-h	1.7a-d	1.6c-g	1.5c-g
B <sub>1</sub>	3.0a-c	3.0a-c	2.7b-g	2.8a-c	2.2b-g	1.7f-i	1.8a-e	1.3d-g	1.5c-g
B <sub>2</sub>	3.0a-c	3.7ab	3.1ab	2.5a-e	2.5a-e	2.5a-e	1.8a-e	2.0a-c	1.5c-g
B <sub>3</sub>	2.9a-d	2.6c-h	2.7b-g	2.8a-c	1.7e-i	2.0d-i	1.8a-e	1.6c-g	1.3d-g
B <sub>4</sub>	2.4e-h	3.0a-e	2.3gh	3.0a	2.6a-d	2.7a-d	1.0g	1.5c-g	1.5c-g
B <sub>5</sub>	2.7b-g	3.0a-c	2.7b-g	2.4a-f	2.6a-d	2.1c-h	2.2ab	1.5c-g	1.3d-g

For each temperature value with different letters show significant difference s determined by DMR Test.

### **Effect of inoculation on plant dry biomass**

In general there was not any pronounced- difference in plant biomass between the two temperatures. Inoculation

of both *Escherichia* and *Bacillus* strains had either insignificant or significantly or negative impact on studies parameter at all glucose regimes at 25 as well as 37 (Table 5).

**TABLE 3 - Effect of bacterial inoculation on root length (cm) of maize var. 6098 at different temperatures (°C) and concentrations of glucose (mg g<sup>-1</sup>)**

Treat.	25°C			37°C			39°C		
	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>
Cont	2.9a-d	3.2a	3.0a-c	2.1c-h	1.3i	1.6g-i	2.0a-c	1.5c-g	1.4c-g
E <sub>1</sub>	2.8a-f	2.8a-f	2.3h	2.4a-f	2.5a-e	2.0d-i	1.9a-d	1.5c-g	1.2c-g
E <sub>2</sub>	3a-c	2.7b-f	3.0a-c	2.6a-d	2.9ab	1.4hi	1.7b-f	1.4c-g	1.3d-g
E <sub>3</sub>	2.4f-h	2.9a-d	2.6d-h	2.2b-g	1.8e-i	2.0f-i	1.1fg	1.3d-g	1.3d-g
E <sub>4</sub>	2.9a-d	3.0a-c	2.8a-f	1.5g-i	1.8e-i	2.1c-h	1.7a-d	1.6c-g	1.5c-g
B <sub>1</sub>	3.0a-c	3.0a-c	2.7b-g	2.8a-c	2.2b-g	1.7f-i	1.8a-e	1.3d-g	1.5c-g
B <sub>2</sub>	3.0a-c	3.7ab	3.1ab	2.5a-e	2.5a-e	2.5a-e	1.8a-e	2.0a-c	1.5c-g
B <sub>3</sub>	2.9a-d	2.6c-h	2.7b-g	2.8a-c	1.7e-i	2.0d-i	1.8a-e	1.6c-g	1.3d-g
B <sub>4</sub>	2.4e-h	3.0a-e	2.3gh	3.0a	2.6a-d	2.7a-d	1.0g	1.5c-g	1.5c-g
B <sub>5</sub>	2.7b-g	3.0a-c	2.7b-g	2.4a-f	2.6a-d	2.1c-h	2.2ab	1.5c-g	1.3d-g

For each temperature value with different letters show significant difference s determined by DMR Test.

**TABLE 4 - Effect of bacterial inoculation on number of roots of maize var. 6098 at different temperatures (°C) and concentrations of glucose (mg g<sup>-1</sup>)**

Treat.	25°C			37°C			39°C		
	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>
Cont	5.7a-c	5.5a-d	3.1i-j	3.3j-m	3.2k-m	3.4j-m	3.7b-f	4a-c	3.5c-h
E <sub>1</sub>	4.3e-h	4.7b-g	3.9g-i	3.8e-l	5.5ab	3.5lm	2.6i-j	2.9g-j	2.5j
E <sub>2</sub>	5.8ab	5.2a-f	4.9b-g	3.9d-l	5.6a	4.5c-g	3.0f-j	2.6i-j	3.1e-j
E <sub>3</sub>	3.6h-j	5.0a-g	3.5h-j	3.3j-m	3.2k-m	4.2c-j	2.7i-j	2.5j	3.3c-i
E <sub>4</sub>	5.7a-c	4.1f-i	4.6c-h	4.0d-l	3.3k-m	4.6c-e	3.2d-j	3.6c-g	4.6ia
B <sub>1</sub>	4.9b-g	4.8b-g	5.5a-d	3.5i-m	3.4g-l	3.7b-d	3.3c-i	3.2e-j	4.4ab
B <sub>2</sub>	5.4a-e	5.7a-c	6.1a	4.1d-k	3.8e-l	4.7b-d	2.6i-j	4.0a-c	2.5j
B <sub>3</sub>	4.5d-h	5.7a-c	4.3e-h	4.4c-i	2.7m	3.6h-l	3.8b-e	3.6c-g	3.0e-j
B <sub>4</sub>	4.3e-h	4.9b-g	4.4f-i	3.7f-l	3.8e-l	4.1c-h	3.1e-j	3.5c-h	3.1e-j
B <sub>5</sub>	2.7i-j	4.1f-i	4.3j	4.4c-i	5.0a-c	4.6c-f	2.8h-j	3.6c-g	3.9a-d

For each temperature value with different letters show significant difference s determined by DMR Test.

**TABLE 5 - Effect of bacterial inoculation on dry weight of maize var. 6098 at different temperatures (°C) and concentrations of glucose (mg g<sup>-1</sup>)**

Treat.	25°C			37°C			39°C		
	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>	0 mg g <sup>-1</sup>	1.25 mg g <sup>-1</sup>	2.5mg g <sup>-1</sup>
Cont	2.0 c	2.0 a-c	2.2 a	1.7 d-e	1.9 b-c	2.5 a	1.5 b	1.8 b	1.9 a
E <sub>1</sub>	1.6 d-e	1.5 e	2.0 a-c	1.6 e-f	1.4 g	1.5 f-g	2.0 a	1.2 b	1.9 a
E <sub>2</sub>	1.2 g-h	1.8 c-d	1.4 e-f	1.5 f-g	1.9 b-c	1.8 c-d	1.7 b	1.9 a	1.4 b
E <sub>3</sub>	1.3 f-g	2.0 a-c	1.8 c-d	1.8 c-d	1.8 c-d	1.8 c-d	2.0 a	1.5 b	1.0 b
E <sub>4</sub>	1.7 e-f	1.8 c-d	1.9 b-c	.62 j	.86 i	1.5 f-g	.70 b	.75 b	.84 b
B <sub>1</sub>	1.3 h	1.8 c-d	1.8 c-d	1.9 b-c	.62 j	.71 j	.88 b	.74 b	.79 b
B <sub>2</sub>	1.9 c-d	1.8 c-d	2.0 a-c	1.7 d-e	1.7 d-e	.66 j	1.4 b	1.8 a	1.4 b
B <sub>3</sub>	1.8 c-d	1.6 de	1.9 b-c	1.9 b-c	1.8 c-d	1.8 c-d	1.9 a	1.9 a	2.0 a
B <sub>4</sub>	1.5 e	2.0 a-c	2.1 a-b	1.2 h	2.0 b	1.5 f-g	1.5 b	1.8 b	1.9 a
B <sub>5</sub>	1.8 c-d	2.2 a	2 a-c	1.4 g	1.8 c-d	2.0b	1.9 a	1.5 b	2.0 a

For each temperature value with different letters show significant difference s determined by DMR Test.

and in plant productivity. They enhance the plant growth by enhancing mineral uptake (14) but also destroy pathogen of plants by release of bacterial metabolite, antibiotics, ammonia and cyanide. Complex bacterial communities are responsible for driving the biogeochemical cycling that maintains biosphere (15, 16, 17). Bacteria either occur singly or in communities. These communities may be comprised of single bacterial spp or of different bacterial species.

Much diverse type of bacterial cells assembled as biofilm communities. Biofilms are broadly defined as assemblage of microorganisms and their associated extracellular products at an interphase, typically attached to a biotic or abiotic surface. The extracellular products are exopolysaccharides (EPS), which are larger components of bacterial biofilm. These exopolysaccharides have significant contribution to biofilm structure and function (18). Various carbon sources that may be potential selective agents for positive selection systems. The rationale behind the experiments within study lies the fact that the plant cannot use or metabolize all carbon sources effectively, and thus these can be used as limiting factors to regeneration, growth and development. Galactose, glucose and fructose (or levulose) are monosaccharide and should be more easily decomposed (19). Carbon metabolite (hexose)-mediated regulatory mechanisms regulate photosynthesis and provide the necessary integration with plant metabolism and the genes encoding them (20). Gene regulation by hexoses occurs when the depletion of sugars results in activation of gene expression and to an increase in photosynthetic capacity. So when glucose was supplemented in soil the response of maize shoot length was variable at various temperature (25, 37 and 39°C). At 25°C increases in shoot length occur at both concentrations (1.25, 2.50 mg g<sup>-1</sup>) but maximum increase at 1.25 mg g<sup>-1</sup> glucose concentration, where as at 37°C with increase of glucose concentration decrease in shoot length occurred. At 39°C increases in shoot length with increase in concentration of glucose. So under non inoculated condition, shoot length showed positive response at 39°C, negative at 37°C and moderate at 25°C. In case of inoculated treatments variable results were obtained. Bacterial inoculation is reported to stimulate plant growth and yield (16, 21, 22, 23). Bacterial inoculation showed either

increase or decrease in shoot length of maize seedling at 25. Some treatments showed increase while other decrease with increase of glucose concentration at different temperatures. E1, B4 treatments increase with increase of glucose concentration, B2, B3 decrease while E2, E4, B1, B5 increase more at 1.25 mg g<sup>-1</sup> glucose concentration, E3 decrease at 1.25 mg g<sup>-1</sup> and increase at 2.50 mg g<sup>-1</sup> glucose concentration. At 37°C E4, B4, B5 increase with increase of glucose concentration, E3, B2 decrease while E3 showed equal increase at both concentration of glucose. E2, B1 more increase at 1.25 mg g<sup>-1</sup>, E1 increase at 1.25 mg g<sup>-1</sup> and decrease at 2.50 mg g<sup>-1</sup> concentration of glucose. At 39°C E1, B1 increase with increase of glucose concentration, E3, B3, B4, B5 decrease while B2 increase at 1.25 mg g<sup>-1</sup> glucose concentration and decrease at 2.50 mg g<sup>-1</sup> glucose concentration. E2 showed no effect at 1.25 mg g<sup>-1</sup> but decrease at 2.50 mg g<sup>-1</sup> glucose concentration. Various carbon sources have pronounced effect on plant growth. Glucose promotes root growth, fructose promotes shoot development and sucrose promotes both shoot development and root growth in asparagus although this was not quantitatively discriminated as in this study. (24). Under non inoculated condition, more increase in root length occurred at 1.25 mg g<sup>-1</sup> at 25°C and 39°C while decrease in root length occurred at 2.50 mg g<sup>-1</sup> glucose concentration at 37°C. So root length show positive response at 1.25 mg g<sup>-1</sup> glucose concentration at 2.50 mg g<sup>-1</sup> and 39°C while negative at 2.50 mg g<sup>-1</sup> glucose concentration at 37°C. As seeds germinate and roots grow through the soil the loss of organic material provides the driving force for the development of active microbial populations around the root, known as the rhizosphere effect (25, 26). The rhizosphere (root surface and region immediately surrounding root) constitute an ecological niche in soil where nutrient are more readily available and certain bacteria have developed mechanism to take advantage of this niche (27). Rhizosphere is highly important source of organic materials in soil it contributes to organic matter directly through the release of soil root material and its decompositions and indirectly through root exudation, which stimulate microbial activity and biomass (28). Bacterial inoculated treatments showed various responses under different conditions. At 25°C B3 increase with

increase of concentration, E2, E3 decreases with increase of glucose concentration, E1, E4, B5 increase at 1.25 mg g<sup>-1</sup> glucose concentration while decrease at 2.50 mg g<sup>-1</sup> glucose concentration, B1 showed equal increase at both concentration of glucose, B2 showed equal decrease at both concentrations, B4 decrease at 1.25 mg g<sup>-1</sup> glucose concentration while decrease at 2.50 mg g<sup>-1</sup> glucose concentration. At 37°C B5 increase with increase of glucose concentration, E1, B2, B3, B4 increase more at 1.25 mg g<sup>-1</sup> glucose concentration than 2.50 mg g<sup>-1</sup> glucose concentration. B1 showed increase at 1.25 glucose concentration and decrease at 2.50 mg g<sup>-1</sup> glucose concentration, E2, E4 decrease at 1.25 mg g<sup>-1</sup> glucose concentration and increase at glucose concentration, E3 decrease more at 1.25 mg g<sup>-1</sup> glucose concentration than 2.50 mg g<sup>-1</sup> glucose concentration. At 39°C root length showed variable response with different treatments. E2, B2, B4 increase at 1.25 mg g<sup>-1</sup> glucose concentration and decrease at 2.50 glucose concentration, E3, B3 decrease with increase of concentration E1 more decrease at 1.25 mg g<sup>-1</sup> than 2.50 mg g<sup>-1</sup> glucose concentration. E4 showed no effect at 1.25 glucose concentration but decrease at 2.50 mg g<sup>-1</sup>, B1 decrease at 1.25 mg g<sup>-1</sup> but increase at 2.50 mg g<sup>-1</sup> glucose concentration. B5 decrease more at 2.50 mg g<sup>-1</sup> glucose concentration. The formation of shoots is highly dependent on the carbon source and on light conditions. Highest shoot formation occurred when sucrose was utilized (19, 24). Various carbon sources that may be potential selective agents for positive selection systems. Various concentrations of glucose under different temperature conditions showed variable effect on number of leaves. Increase in number of leaves was more pronounced at 1.25 glucose concentration at 25°C while more decrease in number of leaves occurred at 1.25 mg g<sup>-1</sup> than 2.50 mg g<sup>-1</sup> at 37°C while decrease in number of leaves occurred with increase of concentration at 39°C. So number of leaves either increase or decrease variably in non inoculated treatments. Under natural conditions, microorganisms obtain essential nutrients required to grow, synthesize and reproduce new cells from their immediate environment. Some bacteria can survive and maintain populations in environments with only micro molar concentrations of organic substrates (29). Number of leaves showed variable growth response under various carbon source

concentrations and temperatures. At 25 °C, B2, E3 increase more at 1.25 mg g<sup>-1</sup> glucose concentration in comparison to 2.50 mg g<sup>-1</sup> glucose concentration, E4, B4 increase at 1.25 mg g<sup>-1</sup> glucose concentration while decrease at 2.50 mg g<sup>-1</sup> glucose concentration. B5 increase at 1.25 mg g<sup>-1</sup> glucose concentration but has no effect at 2.50 mg g<sup>-1</sup> glucose concentration, B3 showed decrease at both glucose concentrations. E2 decrease at 1.25 mg g<sup>-1</sup> glucose concentration while showed no effect at 2.50 glucose concentration, B1, E1 showed no effect on number of leaves at 1.25 mg g<sup>-1</sup> glucose concentration but decrease at 2.50 mg g<sup>-1</sup> glucose concentration. At 37°C E4 increased with increase of concentrations, E1, E2, B5 increase at 1.25 mg g<sup>-1</sup> glucose concentration decreases at 2.5 glucose concentration, B1 decrease with increase of concentrations. E3, B3, B4 showed more decrease at 1.25 glucose concentration than 2.50 glucose concentration. B2 showed no change in number of leaves at both glucose concentrations.

Different treatments showed various responses at 39°C. E3, B4 showed equal increase in number of leaves at both glucose concentrations, B2 showed increase at 1.25 glucose concentration but decrease at 2.50 glucose concentration. B1 showed more decrease at 1.25 mg g<sup>-1</sup> glucose concentration. E1, E2, E4, B3, B5 decrease with increase of concentration. Plant-microbial C and N interactions in the rhizosphere (30). Determined the processes and spatial and temporal patterning by which roots alter mineralization of C and N in the rhizosphere carbon source supplementation caused increase in number of roots with increase of glucose concentration at 39°C, decrease in number of roots occurred with increase of concentration at 25°C while decrease at 1.25 mg g<sup>-1</sup> glucose concentration but increase at 2.50 mg g<sup>-1</sup> glucose concentration. So number of roots showed positive response at 39, negative at 25 while moderate at 37°C. Microbial biofilms are implicated in the natural and modulated environmental cleanliness systems, used for the purification of drinking water, detoxification of oil spills, removal of heavy metals and biodegradation of the hazardous xenobiotics in the contaminated waters and soils. Because of physio chemical and biological properties biofilms are highly beneficial for removing organic and inorganic contaminant from the natural environment and in modulated systems (31). Microbial biofilm formation with the

plant roots protect the plants against soil borne diseases and improves crop productivity by regulating nutrient and water flow across the roots, as well as by improving the physio chemical characteristics of soils. At 25°C B2 showed increase in number of roots with increase of glucose concentration, B3, B4, B5 showed more increase at 1.25 mg g<sup>-1</sup> glucose concentration than 2.50 glucose concentration. E2 decrease with increase of concentration, E4 showed more decrease at 1.25 mg g<sup>-1</sup> glucose concentration, E1, E3 showed more increase at 1.25 mg g<sup>-1</sup> glucose concentration but decrease at 2.50 mg g<sup>-1</sup> glucose concentration, B1 decrease at 1.25 mg g<sup>-1</sup> glucose concentration but increase at 2.50 mg g<sup>-1</sup> glucose concentration. Number of roots varied significantly to various treatments under different glucose concentration at 37°C. B4 showed increase with increase of concentration, E2, B5 showed more increase at 1.25 mg g<sup>-1</sup> glucose concentration. E1 increase at 1.25 mg g<sup>-1</sup> glucose concentration but decrease at 2.50 mg g<sup>-1</sup> glucose concentration. E3, E4, B1, B2 decrease at 1.25 mg g<sup>-1</sup> but increase at 2.50 mg g<sup>-1</sup> glucose concentration. B3 showed more decrease at 1.25 mg g<sup>-1</sup> glucose concentration. At 39°C E4, B5 showed increase with increase of concentration, B2 showed more increase at 1.25 mg g<sup>-1</sup> glucose concentration, B4 showed increase at 1.25 mg g<sup>-1</sup> glucose concentration while no significant effect at 2.50 mg g<sup>-1</sup>. E1 showed increase at 1.25 mg g<sup>-1</sup> glucose concentration. E2, E3, B1 showed decrease at 1.25 mg g<sup>-1</sup> glucose concentration and increase at 2.50 mg g<sup>-1</sup> glucose concentration. B3 decrease with increase concentration Carbon metabolite (hexose)-mediated regulatory mechanisms regulate photosynthesis and provide the necessary integration with plant metabolism and the genes encoding them (20). Gene regulation by hexoses occurs when the depletion of sugars results in activation of gene expression and to an increase in photosynthetic capacity. Dry weight increase with increase of concentration at 37°C, 39°C while at 25°C showed no effect at 1.25 mg g<sup>-1</sup> glucose concentration but increase at 2.50 glucose concentration. So dry weight showed positive response at 37°C and 39°C while moderate at 25°C. Some bacteria can survive and maintain populations in environments with only micro molar concentrations of organic substrates (32). Some bacteria can survive and maintain populations in environments with only micro molar concentrations

of organic substrates (32). Dry weight of maize seedling respond varyingly at different glucose concentration and temperatures. Under 25°C E4, B4 showed increase with increase of concentration, E2, E3, B5 showed more increase at 1.25 mg g<sup>-1</sup> glucose concentration. B2, B3 decrease with increase of concentration, E1 decrease at 1.25 mg g<sup>-1</sup> glucose concentration and increase at 2.50 glucose concentration. B1 showed equal increase at both concentrations. At 37°C E4, B5 increase with increase of concentration, B1, E1 showed decrease with increase of concentration. E2, B4 more increased at 1.25 mg g<sup>-1</sup> glucose concentration. E3 showed no effect on dry weight of seedling at both concentration. B2 showed no effect at 1.25 but decrease at 2.50 mg g<sup>-1</sup> glucose concentration, B3 decrease at both concentrations. At 39°C E4, B4 increase with increase of concentration, E1, E3, B1 showed decrease with increase of concentration. E2 increase at 1.25 mg g<sup>-1</sup> glucose concentration but decrease at 2.50 mg g<sup>-1</sup> glucose concentration. B2 increase at 1.25 mg g<sup>-1</sup> glucose concentration but showed no effect at 2.50 mg g<sup>-1</sup> glucose concentration, B3 showed no effect at 1.25 mg g<sup>-1</sup> glucose concentration but increased at 2.50 mg g<sup>-1</sup> glucose concentration. B5 decreased at 1.25 mg g<sup>-1</sup> and increase at 2.50 mg g<sup>-1</sup> glucose concentration.

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