



Effect of Different Concentrations of Garlic Extract on Crown Gall and Seedling Growth of Some Varieties of Tomato (*Lycopersicon Esculentum* Mill.) Grown in Dutse Town, Jigawa State

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ABSTRACT

Effect of different concentrations of garlic extract on crown gall of some varieties of tomato (*lycopersicon esculentum*) was investigated. Crown gall is a plant disease caused by the soil bacterium *Agrobacterium tumefaciens*. The experiment was conducted at Agricultural farm Federal University Dutse, Jigawa State. (Latitude 11.000N to 13.00N and longitude 8.000E to 10.50E). The duration of the study was between August to October, 2019. Two varieties of tomato plants were used, a local variety and an improved variety which are normally grown in Dutse town. Plant infected with crown gall as a source of *A. tumefaciens* bacteria were obtained from experts at local nursery and "stab inoculation" technique using a pin was used to infect the tomato seedlings with the bacteria. The experimental design used was completely randomized block design. Different concentrations of garlic extract were used at; 100%, 75%, 50%, 25% concentration levels and then the control. Data was collected on number of crown galls for each of the two varieties and growth parameters such as plant height, number of leaves, fresh weight and dry weight. From the result 50% concentration of the garlic extract had more effect on the crown gall than any of the treatments, followed by 75% then 25% then 100% garlic extract. With regards to the two types of varieties after the inoculation of the plants with the bacteria, the improved variety had the least number of infected plants (90%) and responded well to the treatment and had the highest seedling growth more than the local variety that had the highest number of infected plants (98%) and had reduced seedling growth. The implication and significant of this findings is that elimination of crown gall and high seedling growth were obtained using 50% garlic concentration. This shows that high garlic concentration (100%) has negative effect on plant growth and therefore reduced seedling growth due to its high concentration. This indicates that moderate concentration (50%) is needed to combat the disease and allow growth. Therefore selecting such concentration over any other will be a wiser choice as it is more effective in tackling crown gall disease.

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INTRODUCTION

Agrobacterium tumefaciens is the causal agent of crown gall disease. Crown gall is a plant disease that affects a large variety of broad-leaved (dicotyledonous) plants, including tomatoes, apple, pear, cherry, almond, raspberry,

and rose plants leading to several losses. With the ability to infect many different plant species, *A. tumefaciens* possibly has the broadest host range as compared to other plants pathogenic bacterium. *Agrobacterium tumefaciens* cleverly transfers a genetic principle to plant host cells and

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integrates it into their chromosomes. (Clarence, 2014).

Crown gall disease is particularly caused by the bacteria *Agrobacterium tumefaciens* and antibiotics cannot be used for agricultural purposes, because resistant bacterial strains will develop with widespread use, resulting eventually in loss of effectiveness of the antibiotic. However, garlic a natural product has been shown to have anti-bacterial properties under some conditions (Jonkers et al., 1999). Hence its selection for the experiment.

Garlic (*Allium sativum*) is a species in the onion genus, *Allium*. A perennial herb with flat, fleshy greyish leaves and a bulb, composed of typical garlic cloves. Its close relatives include the onion, shallot, leek, chive, and Chinese onion (Block and Eric, 2010)

Garlic is native to Central Asia and northeastern Iran, and has long been a common seasoning worldwide, with a history of several thousand years of human consumption and use. (John, 2001)

Allium sativum is a bulbous plant, growing up to 1 metre (3.3 ft) in height. It produces hermaphrodite flowers. It is pollinated by bees, butterflies, moths, and other insects. (Meredith et al., 2014).

The tomato is the edible, often red, berry of the plant *Solanum lycopersicum*, commonly known as a tomato plant. The species originated in western South America and Central America. The Nahuatl (Aztec language) word *tomatl* gave rise to the Spanish word *tomate*, from which the English word *tomato* derived. Its domestication and use as a cultivated food may have originated with the indigenous peoples of Mexico. Tomatoes are a significant source of umami flavor. The tomato is consumed in diverse ways, raw or cooked, in many dishes, sauces, salads, and drinks. While tomatoes are fruits—botanically classified as berries—they are commonly used as a vegetable ingredient or side dish. (Fleming, 2013).

Aims and Objectives

The aim of this experiment is to evaluate the effect of different concentrations of garlic extract on Crown Gall of some varieties of tomato

grown in Dutse Town. While the objectives were To determine the effect of garlic extract on crown gall of some varieties of tomatoes grown in Dutse town, to assess the effect of garlic extract on seedling growth of tomatoes and to determine the best concentration that will inhibit the growth of the bacterium.

MATERIALS AND METHODS

Study site

The experiment was conducted at Agricultural farm Federal University Dutse, Jigawa State found in Sudan savannah ecological zone and lie between latitude 11.00° N to 13.00° N and longitude 8.00° E to 10.15° E. Jigawa is characterized by two seasons, the rainy and the dry seasons. The rainy season usually begins from May and ends in September with heavier rainfall in July and August, October to May marks the dry season with little or no rainfall. Mean temperature during the study period was around 35°C maximum temperature and 28°C minimum temperature.

Source of Seed

The seeds were sourced from Jigawa State Agricultural and Rural Development Authority (JARDA), Dutse head office, Jigawa state.

Source of Garlic Extract

Garlic bulb were sourced from Dutse ultra-modern market. The garlic bulb were crushed and put into a beaker, 100ml of methanol was poured into the beaker and allowed for 24hours. The beaker was then inserted into a water bath at 39°C until the methanol evaporates.

Source of the Disease

The affected seedlings were sourced from expert at local nursery.

Experimental Design

The experimental design used was completely randomized block design (CRBD) and 3 replicates were used, two varieties of the tomato seedlings were used. Plant infected with crown gall as a source of *A. tumefaciens* bacteria were obtained from experts at local nursery and "stab inoculation" technique using a pin were used to

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infect the tomato seedlings with the bacteria after the crown gall was cut-open.

Soil Preparation

The collected soil were air dried under the sun, polythene bags were filled with $\frac{3}{4}$ of its length with prepared soil.

Preparation of Extracts

Cloves of garlic were collected and were air dried at room temperature for two weeks. The dried clove were ground into fine powder using a mortar and pestle. 40 g of the powder was weighed and soaked in 1000 ml of distilled water for 24 hours. The solution was filtered through double layer of muslin cloth followed by No.1 Whatman filter paper. Five concentration levels were prepared i.e 25%, 50%, 75%, and 100% and stored separately in conical flasks. Distilled water was used as control (0 %) as described by (Jonkers et al., 1999).

Planting

Polythene bags (25x20) cm were used for the experiment. The bags were perforated at the lower end to allow free drainage of water, and then filled with soil mixture and placed in a sunken bed. Later they were labeled and placed in completely randomized block design. All seeds were treated and sown directly in the prepaid soil. There were 15 replications for each treatment. The planting was done on 12th August 2019.

Experimental Treatments

The treatment used for the experiment was garlic extract at 25%, 50%, 75% and 100% concentration levels and then the control (water). The infected seedlings were treated with 1 mL of the garlic solution over the stab wound immediately after inoculation, and then once a day for four days. While the control were treated with 1 ml of water over the stab wound immediately after inoculation, and then once a day for four days. The size of the experimental unit was 6x4.8m. There were eight plots of sizes 3x1.2m, four plots for local variety and four plots for improved variety with three replications. There were fifteen plants in each plot with a total plant population of 120. Data was collected every week for seven weeks.

Data collection

The data was collected on the following growth parameters:

Plant Height in (cm)

The height of the control and treated plants were measured weekly from the ground level to the tip of the stem or terminal bud with the aid of a meter rule and thread and the measurements were recorded.

Number of Leaves

The total number of leaves on each of the treated and control plants were counted weekly and the mean was recorded for each treatment.

Plant Fresh Weight: Here the weights of the plants were recorded immediately after they were harvested.

Plant Dry Weight: Here the weight was recorded after drying the plant at temperature higher than the ambient air temperature (around 65-100°C) to drive off the water.

Data Analysis

The data collected was subjected to one-way analysis of variance (ANOVA) and significant differences were further treated to a least significant difference test at $P = 0.05$

RESULT AND DISCUSSION

Percentage Number of Crown Galls

The result of the different varieties and treatments on the number of crown galls is shown in Table 1. Comparison of the varieties showed that there was significant difference ($P \leq 0.05$) between the two varieties, after the inoculation of the plants with the bacteria the local variety were more affected and had the highest number of crown gall than the improved variety with 98% and 90% for local and improved varieties respectively while between the treatments the control plants were more affected with the crown gall than any other. This was followed by 25% then 100% then 75%, the least in the number of crown galls were those plants treated with 50% garlic extract (Table 2).

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Table 1: Comparison between the tomato varieties on number of seedlings with crown gall

Variety	percentage number of crown gall (%)
Improved	90
Local	98
Mean	94.0
LSD (0.05%)	3.04

Table 2: Effect of different concentrations of garlic extract on number of crown galls at different weeks after planting

Treatment	Wk1	Wk2	Wk3	Wk4	Wk5	Wk6	Wk7
100%	7	12	10	10	8	7	5
75%	4	9	8	7	5	3	2
50%	5	7	7	5	3	1	0
25%	7	7		5	5	4	4
0%	7	9	10	10	12	13	14
Mean	6	8	8	7	6	6	4
LSD (0.05%)	0.20	0.18	0.17	0.39	0.51	0.11	0.10

Seedling growth parameters:

Plant Height

Comparison between the treatments on plant height of the improved and local varieties of tomato

The effect of garlic extract on crown gall of tomato was tested using five different concentrations (100%, 75%, 50%, 25% and 0%) on two varieties of tomato (Improved and local varieties). From the result obtained 100% concentration had reduced plant height more than any of the treatments. This showed that the higher the extract concentration the more the reduction in height and vice versa. The control plants had stunted growth throughout as they were infected with the bacteria and did not receive the garlic treatment.

Comparison between the treatments on the plant height of the improved variety showed that those plants treated with 50% garlic extract had the highest plant height in all the weeks i.e at 1,2,3,4,5, 6 and 7 weeks after planting with 0.98, 3.03, 5.87, 11.88, 17.58, 22.58, 27.33cm respectively. Followed by those plants treated with 75% concentration extract with (1.05, 2.95, 5.43, 9.51, 14.08, 16.8, 23.03cm) at 1, 2,3,4,5,6 weeks after planting respectively, this was followed by 25% treatment (0.98, 2.97, 5.67, 10.08, 14.38, 17.33, 23.78cm) then 100% (1.03,3.08, 5.83, 10.2cm, 13.87, 15.76, 19.08cm) the least in plant height were the control plants with 1.02, 2.98,

5.63, 8.82, 9.58, 10.33, 11.13cm at 1,2,3,4,5, 6 weeks after planting respectively. The difference between all the treatments was significant. (Table 3).

Comparison between the varieties on plant height

Comparison between the varieties on plant height showed that the improved variety had the highest plant height (14.8, 16.31, 6.9, 17.4, 18.0, 18.7, 9.8cm) than the local variety (8.4, 11.0, 12.4, 14.5, 14.08, 16.0, 16.8cm) in all the weeks. (Table 4). This was because the improved variety responded well with the treatment more than the local variety. Block and Eric, 2010 reported that Fresh garlic is an effective remedy against infections because of its anti-bacterial and anti-viral properties. When raw garlic is crushed a strong oxidant allicin is formed which has strong antibiotic property that kill many types of bacteria.

Garlic extract may have variable effects, based on different concentrations as (Rutledge *et al.*,2014) observed greater effects of the fresh garlic extract on crown gall infection and growth of some seedlings of Raspberry under medium concentration. Similarly Meredith *et al.*, 2014 reported that high concentration has harmful effects on germination and seedling growth of wheat, barley, lentil, chickpeas and mustard infected with bacterial disease.

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Agarry *et al*, 2005 reported that the garlic extract was potent against three strains of Mycobacterium (*M. fortuitum*, *M. smegmatis* and *M. kansasii*) and a strong antimycobacterial activity against *M. tuberculosis* as well as antibacterial activity against *P. aeruginosa*, *E. coli*, *S. aureus* and *S. typhi*. The

preliminary phytochemistry revealed presence of terpenoids, flavonoids and tannins. Thus, *garlic* could be a rich source of antimicrobial agents and it can give scientific backing to its use by the local people of Lake Victoria region of Kenyas. (Mothana and Linclequist, 2005).

Table 3: Effect of different concentrations of garlic extract on plant height at different weeks after planting.

Garlic extracts conc.	Wk1	Wk2	Wk3	Wk4	Wk5	Wk6	Wk7
100%	1.03	3.08	5.83	10.2	13.87	15.76	19.08
75%	1.05	2.95	5.43	9.51	14.08	16.8	23.03
50%	0.98	3.03	5.87	11.88	17.58	22.58	27.33
25%	0.98	2.97	5.67	10.08	14.38	17.33	23.78
0%	1.02	2.98	5.63	8.82	9.58	10.33	11.13
Mean	1.00	3.00	5.68	10.09	13.89	16.56	20.87
LSD (0.05%)	0.08	0.05	0.42	0.49	0.53	0.96	1.32
Variety							
Improved	14.8	16.3	16.9	17.4	18.0	18.7	19.8
Local	8.4	11.0	12.4	14.5	14.08	16.0	16.8
Mean	11.6	13.7	14.7	15.9	16.0	17.4	18.3
LSD (0.05%)	0.33	0.38	0.42	0.44	0.48	1.22	1.28

Number of Leaves

Comparison between the treatments on number of leaves of the improved variety of tomato

Comparison between the treatments on number of leaves of improved variety showed that those plants treated with 50% garlic extract had the highest number of leaves with 2,9, 15, 19, 21, 24,27 at at 1,2,3,4,5,6 weeks after planting respectively. The difference between all the treatments was significant.(Table 4). This was followed by 75% then 25% and then the 100% concentration. The least with the number of leaves were the control plants.

Comparison between the two varieties showed that the improved variety had the highest number of leaves (7, 9, 12, 15, 16, 19 23) than the local variety (4, 7, 10, 12, 14, 15 19) in all the weeks and the difference between them was significant.(Table 4). This is because Meredith *et al.*, 2014 reported that the bacteria which causes the crown gall usually prevent the passage of water and nutrient by clogging the affected vessels ceased to function properly this brings about stunted growth and also few leaves formation in the affected plants.

Table 4: Effect of different concentrations of garlic extract on number of leaves at different weeks after planting.

Garlic extracts conc.	Wk1	Wk2	Wk3	Wk4	Wk5	Wk6	Wk7
100%	5	7	9	12	14	15	17
75%	5	9	13	17	19	20	24
50%	6	9	15	19	21	24	27

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25%	4	6	8	11	13	16	18
0%	2	2	3	3	5	6	8
Mean	5	7	10	13	15	17	19
LSD (0.05%)	2.7	0.8	1.0	1.8	2.0	2.3	2.7
Variety							
Improved	7	9	12	15	16	19	23
Local	4	7	10	12	14	15	19
Mean	6	8	11	14	15	17	21
LSD (0.05%)	3.4	0.6	0.8	2.3	2.7	3.0	3.3

Table 5: Effect of different concentrations of garlic extract on fresh weight at different weeks after planting (g).

Garlic extracts conc.	Wk1	Wk2	Wk3	Wk4	Wk5	Wk6	Wk7
100%	0.8	1.5	2.0	2.3	2.5	2.8	3.0
75%	1.0	1.7	2.5	2.9	3.5	4.0	4.9
50%	1.5	1.8	2.7	3.7	5.0	5.5	6.0
25%	0.6	1.0	1.8	2.0	2.5	3.0	3.8
0%	0.3	0.8	1.0	1.5	1.7	1.9	2.0
Mean	0.8	1.4	2.0	2.5	3.2	3.6	4.0
LSD (0.05%)	0.13	0.09	0.07	0.17	0.05	0.04	0.10
Variety							
Improved	1.7	2.8	3.4	4.5	4.9	5.5	6.3
Local	1.1	2.0	2.6	3.6	4.2	4.9	5.4
Mean	1.4	2.4	3.0	4.1	4.6	5.2	5.9
LSD (0.05%)	0.15	0.03	0.09	0.05	0.21	0.04	0.07

Fresh and Dry Weight

The results of fresh and dry weight are presented in table 5 and 6 for Plant fresh weight and Plant dry weight respectively. The treatments induced different responses for Plant fresh weight and Plant dry weight. Between the treatments 50% had the highest number of fresh weight and dry weight with 1.5, 1.8, 2.7, 3.7, 5.0, 5.5, 6.0g and 0.7, 0.7, 0.9, 1.4, 1.7, 1.9, 2.0g for fresh and dry weight respectively followed by 75% with 1.0g, 1.7g, 2.5g, 2.9g, 3.5g, 4.0g, 4.9g and 0.5g, 0.6g, 0.8g, 0.8g, 1.2g, 1.5g 1.8g for fresh and dry weight respectively followed by 25% with 0.6g, 1.0g, 1.8g, 2.0g, 2.5g, 3.0g, 3.8g and 0.3g, 0.3g, 0.5g, 0.7g, 0.9g, 1.0g and 1.1g for fresh and dry weight followed by 100% with 0.8g, 1.5g, 2.0g, 2.3g, 2.5g, 2.8g, 3.0g and 0.4g, 0.5g, 0.6g, 0.6g, 0.7g, 0.7g and 0.9g for fresh and dry weight respectively the least in fresh and dry weight were the control plants. 0.3g, 0.8g, 1.0g, 1.5g, 1.7g, 1.9g, 2.0g and

0.1g, 0.2g, 0.3g, 0.3g, 0.4g, 0.4g, 0.7g for fresh and dry weight respectively.

Comparison between the varieties is also presented in table 5 and 6 and the result showed that the improved variety had the highest Plant fresh weight and Plant dry weight than the local variety and the difference was significant ($P < 0.05$). Genin, 2010 reported similar case with *Ralstonia solanacearum* which is a soil-borne pathogen that infects plants via wounds, root tips or cracks at the sites of lateral root emergence. The bacterium subsequently colonizes the root cortex, invades the xylem vessels and reaches the stem and aerial parts of the plant through the vascular system. *Ralstonia solanacearum* can rapidly multiply in the xylem up to very high cell densities, leading to wilting symptoms and plant death which leads to poor seedling growth including fresh and dry weight of the plant due to the mass death of the plants.

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Table 6: Effect of different concentrations of garlic extract on dry weight at different weeks after planting

Garlic extracts conc.	Wk1	Wk2	Wk3	Wk4	Wk5	Wk6	Wk7
100%	0.4	0.5	0.6	0.6	0.7	0.7	0.9
75%	0.5	0.6	0.8	0.8	1.2	1.5	1.8
50%	0.7	0.7	0.9	1.4	1.7	1.9	2.0
25%	0.3	0.3	0.5	0.7	0.9	1.0	1.1
0%	0.1	0.2	0.3	0.3	0.4	0.4	0.7
Mean	0.4	0.5	0.7	0.8	1.0	1.2	1.7
LSD (0.05%)	0.03	0.06	0.07	0.09	0.05	0.1	0.13
Variety							
Improved	1.7	2.8	3.4	4.5	4.9	5.5	6.3
Local	1.1	2.0	2.6	3.6	4.2	4.9	5.4
Mean	1.4	2.4	3.0	4.1	4.6	5.2	5.9
LSD (0.05%)	0.12	0.16	0.19	0.89	0.21	0.41	0.17

CONCLUSION

In conclusion, garlic extract is found to be effective on tomato crown gall in both improved and local variety only the concentration matters. This is because 50% garlic extract concentration was effective in eliminating the crown galls and increased seedling growth in improved variety while in the local variety 75% concentration was more effective. 100% concentration was found to eliminate the galls but the seedling growth was negatively affected due to the high concentration of the garlic extract. Therefore, moderate concentration is advocated to cure the disease and to allow the plant to grow well.

RECOMMENDATIONS

Some of the recommendations are listed below:

1. Since garlic extract at 50% concentration and 75% were effective in eliminating the crown galls in improved variety and local variety respectively, therefore these concentrations should be used.
2. The garlic extract should be tested on other diseases of plants as it has antibacterial and antifungal properties.
3. Testing a natural product as an antibacterial agent such as Garlic extract that has been shown to have antibacterial properties should be used instead of antibiotics because resistant bacterial strains will develop with widespread use, resulting eventually in

loss of effectiveness of the antibiotic against the disease.

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