International Baccalaureate

Extended essay cover

SAMPLE C

Diploma Programme subject in which this extended essay is registered: Biology

(For an extended essay in the area of languages, state the language and whether it is group 1 or group 2.)

Title of the extended essay: Do the plant extracts of *Allium sativum* (Garlic) and *Catharanthus roseus* (*Vinca rosea*) reduce the growth of *Bacillus subtilis* and *Escherichia coli* and do they show similar effect as antibiotics (Gentamicin)?

Candidate's declaration

If this declaration is not signed by the candidate the extended essay will not be assessed.

The extended essay I am submitting is my own work (apart from guidance allowed by the International Baccalaureate).

I have acknowledged each use of the words, graphics or ideas of another person, whether written, oral or visual.

I am aware that the word limit for all extended essays is 4000 words and that examiners are not required to read beyond this limit.

This is the final version of my extended essay.

Candidate's signature: _____ Date: 04.03.2009

IB Cardiff use only: A: 4638/ B: _____

Supervisor's report

The supervisor must complete the report below and then give the final version of the extended essay, with this cover attached, to the Diploma Programme coordinator. The supervisor must sign this report; otherwise the extended essay will not be assessed and may be returned to the school.

Name of supervisor (CAPITAL letters) ___

Comments

Please comment, as appropriate, on the candidate's performance, the context in which the candidate undertook the research for the extended essay, any difficulties encountered and how these were overcome (see page 13 of the extended essay guide). The concluding interview (viva voce) may provide useful information. These comments can help the examiner award a level for criterion K (holistic judgment). Do not comment on any adverse personal circumstances that may have affected the candidate. If the amount of time spent with the candidate was zero, you must explain this, in particular how it was then possible to authenticate the essay as the candidate's own work. You may attach an additional sheet if there is insufficient space here.

took the initiative to do the pilot study to check the feasibility of the topic during the summer vacation and was ready with all the background work ahead of schedule.

Originally, she did all her experiment in the school lab, but was not able to achieve the level of sterility required to get accurate readings, as the school did not have a laminar airflow chamber. So, the experiment was repeated in a nearby research lab which had a laminar air flow chamber.

Throughout the research, has impressed me with her level of commitment, ability to work methodically and the maturity with which she has handled all situations.

I have read the final version of the extended essay that will be submitted to the examiner.

To the best of my knowledge, the extended essay is the authentic work of the candidate.

I spent [0hrs] hours with the candidate discussing the progress of the extended essay.

Supervisor's signature: _____ Date: 6th March '09

There is an enormous amount of data collected from the experiments. It is a very thorough investigation and well planned. Very little problems have emerged. ✓

INTERNATIONAL BACCALAUREATE
EXTENDED ESSAY
BIOLOGY

TOPIC: To study the antimicrobial property of different plant extracts

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RESEARCH QUESTION: Do the plant extracts of *Allium sativum*(Garlic) and *Catharanthus roseus*(Vinca rosea) reduce the growth of *Bacillus subtilis* and *Escherchia coli* and do they show a similar effect as antibiotics(Gentamicin)?

There is some confusion about the control.
Did you use acetone or Ethanol. ?

Use of acetone would explain the absence of a zone of inhibition as it probably all evaporated before the discs were placed.

This would of course also invalidate a lot of what you have written.

Candidate name:

Candidate number: ✓

School name:

School code:

Session: May 2009

Word count: 3806 ✓

/

Session: May 2009

Acknowledgements:

First, I would like to thank for her support and guidance. I would also like to thank my for letting me use the Biology laboratory for my entire experiment. I would also like to thank for all her encouragement. In addition, I would like to thank for his constant support.

Session: May 2009

ABSTRACT:

An experiment was conducted to inspect the antimicrobial effects of plant extracts. The research question was thus formulated as: **Do the plant extracts of *Allium sativum* (Garlic) and *Catharanthus roseus* (Vinca rosea) reduce the growth of *Bacillus subtilis* and *E.coli* and do they have a similar effect as the antibiotic Gentamicin?** RQ

The extracts of *Allium sativum* and *Catharanthus roseus* were prepared in 100% ethanol using a pestle and mortar. The cultures were grown in a nutrient broth after which they were introduced to the Petri dishes using micropipettes. Paper discs soaked with different concentrations of the plant extracts were placed on the bacterial cultures spread in the Petri dishes and the set up was incubated at 37°C overnight. Readymade Gentamicin discs were also placed along with this. The zone of inhibition around each paper disc was measured the next day using a ruler. The data collected was subjected to two-tailed t-test under 98 degree of freedom.

The comparison of the 't' values of the extracts of Garlic and Vinca rosea with the control resulted in a higher value than the table value at 98 degree of freedom.The hypothesis was accepted that the extracts reduced the growth of *E.coli* and *Bacillus subtilis*. Comparison of the 't' values of the extracts of Vinca rosea and Garlic with Gentamicin resulted in a 't' value lower than table value at 98 degree of freedom so the hypothesis was accepted that there was no difference in the effect shown between Gentamicin and the extracts on both the cultures.

The results indicated that both extracts of Garlic and Vinca rosea reduced the growth of *Escherchia coli* and *Bacillus subtilis* and that the plant extracts showed a same effect as the antibiotic Gentamicin. Also, both the plant extracts had a similar antimicrobial effect on both the cultures. Conclusion -

WORD COUNT: 285 ✓

* Does the ethanol have an effect by itself? I hope a control of ethanol is used. ✓

✓

Session: May 2009

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✓

Need annexe.

LIST OF FIGURES:

Check + correct.

Session, May 2009

CHAPTER 1: INTRODUCTION

1.1 Research question:

Do the plant extracts of *Allium sativum (*Garlic) and *Catharanthus roseus* (Vinca rosea) reduce the growth of *Bacillus subtilis* and *Escherchia* coli and do they show a similar effect as the antibiotic Gentamicin?

1.2 Genesis:

Infectious diseases have moved up from the 5th position in 1981 to the 3rd position in 1992 as the leading cause for death- an increase by 58%.[1] As diseases are spreading increasingly in today's world, people are treated with numerous allopathic medicines that sometimes have detrimental side effects. Having lived in India all my life where alternate forms of medicine like Ayurveda[2] are very much encouraged, I began to contemplate on the various alternatives to allopathic medicines. After some research, I realized that in such forms of medicine plant extracts are widely used. Certain plant extracts have shown to be effective even on serious diseases like Green tea constituent Epigallocatechin-3-Gallate[3] that arrests carcinoma cells by causing cell apoptosis and cell cycle arrest in them. Also, I learnt that according to WHO about 80% of individuals in developed countries use traditional medicines which contain plant extracts and other compounds derived from medicinal plants.[4] Therefore a thorough study of the efficacy of plant extracts on microbes should be done.

1.3 Pilot survey:

During the summer of 2008, I performed a pilot survey prior to the main experiment at the Tamil Nadu Agricultural University, Coimbatore under the supervision of Professor Dr. Balachander The experiment was performed to test the antimicrobial effects of the extracts of Neem (*Azadirachta indica)*, Onion (*Allium cepa*), Garlic (*Allium sativum*), *Gymnema sylvestene*, *Ocimum basiliuma, Catharanthus roseus* (Vinca Rosea) on the growth of the bacterial cultures of *E.coli* and *Bacillus subtilis.*

---

[1] Maurice M. Iwu, New Antimicrobials of Plant Origin, p 457

[2] 'Ayurveda is a traditional system of Hindu medicine which is based on the idea of balance in bodily systems and uses diet, herbal treatment and yogic breathing'

[3] Journal of the National Cancer Institute vol. 89, No. 24- Dec. 17 1997, p 1881-1886.

[4] Prusti, A , Antibacterial Activity of Some Indian Medicinal Plants, Ethnobotanical Leaflets 12, Issued 18 April 2008, pp227-230

The results of the experiment were:

Escherichia coli

Antimicrobial Agents	**Plate I(measurement of the zone of inhibition)**	**Plate II (measurement of the zone of inhibition)**
Ampicillin	34 mm	34 mm
Gentamicin	30 mm	30 mm
Azardirachta indica	Nil	Nil
Allium cepa	Nil	Nil
Allium sativum	24 mm	25 mm
Gymnema sylvestne	Nil	Nil
Oscimum basiliuma	Nil	Nil
Catharanthus roseus	25 mm	25 mm
Ethanol	Nil	Nil
Disc	Nil	Nil

= 0.0 ?

✓

✓

Bacillus subtilis.

Antimicrobial Agents	**Plate I (measurement of the zone of inhibition)**	**Plate II (measurement of the zone of inhibition)**
Ampicillin	30 mm	30 mm
Gentamicin	28 mm	26 mm
Azardirachta indica	09 mm	07 mm
Allium cepa	Nil	Nil
Allium sativum	24 mm	22 mm
Gymnema sylvestne	Nil	Nil
Oscimum basiliuma	Nil	Nil
Catharanthus roseus	24 mm	24 mm
Ethanol	Nil	Nil
Disc	Nil	Nil

Disc size : 6mm

CONTROLS: Ampicillin and Gentamicin were the antibiotics used. Ethanol and the disc were used as controls.

From the above results, it can be noted that *Allium sativum* and *Catharanthus roseus* had a profound antimicrobial effect on the cultures of *E.coli* and *B.subtilis*. *Azadirachta indica* (Neem), however has an effect on *B.subtilis* but not on *E.coli.* This result is similar to that stated in the article 'Anti microbial effects of some medicinal plants against some gram positive, gram negative and Fungi' written by Sanaa O. Yagoub. This might be due to presence of triterpenoids, phenolic compounds , Carotenoids , steroids , valavinoids, ketones and tetratriterpenoids azadirachtin.

As the extracts of Allium sativum and Vinca rosea appeared to be the most effective, they were selected for the main experiment.

1.4 Theoretical Basis:

GARLIC:

The Garlic extract has a profound antimicrobial effect. This property belongs to Garlic's constituent-Allicen. According to the recent results published by UCLA[5], Kyloic Aged Garlic extract showed inhibition of coronary calcification in patients suffering from diseases related to the coronary artery. Garlic belongs to the family *Alliaceae*[6]. It is grown primarily in China (77% of world's total output) followed by India (4.1%)[7]. The composition of Garlic bulbs is water= 84.09%, organic matter=13.38%, and inorganic matter = 1.53%[8]. It is more effective on gram negative bacteria than Penicillin[9]. It was also termed as "a biological response modifier" by Dr. Benjamin Lau of Loma Linda University school of Medicine in 1992. He found that the sulphur compounds that are present in Garlic help to boost the activity of T-lymphocytes and other immune system components.

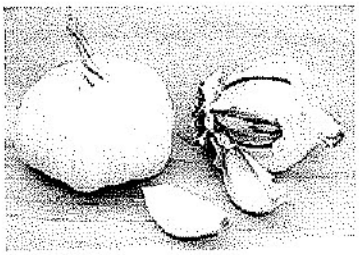

Figure 1- [10] *Allium sativum*

***Catharanthus roseus*:**

Catharantus roseus, formerly called Vinca rosea is a shrubby short-lived plant with oblong leaves and with white or pink flowers. It belongs to the Apocynaceae family. Several antileukamic alkaloids, vinblatine and vincristine were obtained[11]. The extract of Vinca rosea has proven to cure a number of diseases including diabetes. More than seventy alkaloids have been extracted from the roots, stem, leaves and flowers of this plant most of which are monoterpene indole alkaloids.[12] Some of the

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[5] Herbal gram, volume three,p199

[6] Microsoft Encarta Encyclopaedia standard 2007

[7] Wikipedia.com

[8] *Trans. Hon. Soc. Loud.*, new ser., iii. p. 60

[9] Herballegacy.com

[10] Microsoft Encarta Encyclopaedia standard 2007

[11] J. Janick, Perspectives on new crops and new uses, 1999, (Alexandria, VA: ASHS Press),p 457-458

[12] International journal of green pharmacy; July – September 2008; pp 176- 177

alkaloids that were extracted from this plant have shown to exhibit hypoglycemic, tumor inhibiting properties. They have also been used to treat Hodgkin's disease [13]

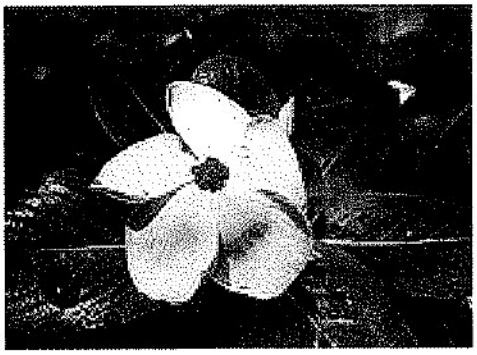

FIGURE 2 – *Catharanthus roseus* [14]

The two organisms used in the experiment are: *Bacillus subtilis* isolated from soil and *Escherichia coli (DH5 alpha)*.

Bacillus subtilis:

Bacillus subtilis belongs to the genus *Bacillus*. It is commonly found in the soil and it is a gram positive bacteria. It forms endospores thereby helping it to survive in tough environmental conditions. This type of Bacteria normally causes food contamination. *B.subtilis* is widely used as a laboratory model and can be considered as a gram positive equivalent to *Escherichia coli*.

[13] Alam, Muzaffar, ISOLATION AND STRUCTURAL STUDIES ON THE CHEMICAL CONSTITUENTS OF CATHARATHUS ROSEUS AND BUXES SPECIES, PhD thesis, University of Karachi, Karachi 1991.

[14] http://en.wikipedia.org/wiki/Rosy_Periwinkle. 4:49PM. 2/16/2009

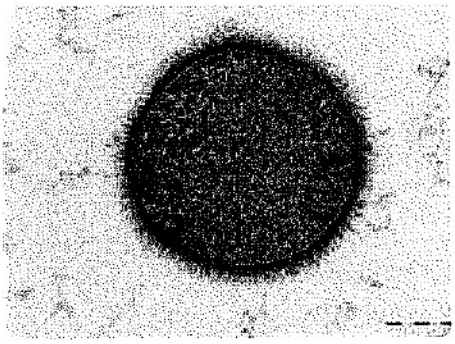

Figure 3 -*Bacillus subtilis*[15]

Escherichia coli:

E.coli belongs to the family Enterobacteriaceae. Most *E.coli* strands are harmless but certain virulent strains can cause Gastroentritis, neo-natal meningitis, urinary tract infection in humans. They are primarily confined to the intestine even though they have the ability to survive outside it for brief periods of time. *E.coli* is a gram negative bacterium.

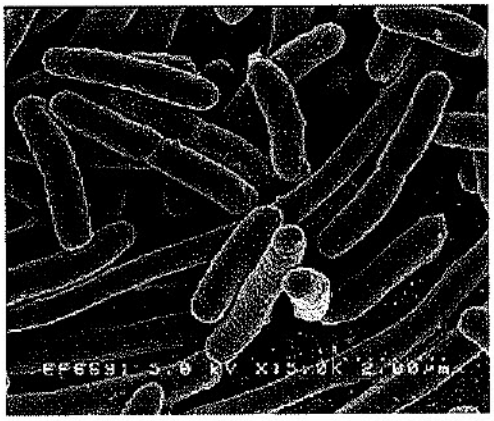

Figure 4 - *E.coli*[16]

[15] http://en.wikipedia.org/wiki/Bacillus_subtilis.4.47pm . 2/16/2009

[16] http://en.wikipedia.org/wiki/Escherichia_coli.4.44pm. 2/16/2009

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Gentamicin[17]:

It is a bactericidal antibiotic that which works by disrupting protein synthesis . It is an aminoglyciside antibiotic synthesised primarily by a gram-positive bacteria.

Academic content is well established but ---

--- presentation is a bit disjointed. Difficult to see a coherent argument up to this point.

[17] Wikipedia.com, 27/2/2009,9.30am

CHAPTER 2: METHODOLOGY

2.1 Objectives of study:

1. To verify if the plant extracts reduce the growth of *Escherichia coli* and *Bacillus subtilis.*
2. To compare antimicrobial action of the plant extract with Gentamicin
3. To compare the antimicrobial activity between the extracts of *Catharanthus roseus* and *Allium sativum*

2.2 HYPOTHESIS

HYPOTHESIS 1: The extracts of *Catharanthus roseus* and *Allium sativum* will reduce growth of on the cultures of *E.coli* and *Bacillus subtilis*

HYPOTHESIS 2: The plant extracts of *Catharanthus roseus* and *Allium sativum* will show a similar antimicrobial effect as the antibiotic Gentamicin

HYPOTHESIS 3: The extracts of *Catharanthus roseus* and *Allium sativum* will show similar antimicrobial effects.

2.3 Variables:

CONSTANT:

1. Temperature (Petri dishes with the cultures were incubated at 37° C)
2. Bacterial cultures

MANIPULATIVE:

1. Concentration of Plant extract (10µl and 15 µl)

RESPONDING:

1. Growth of bacteria

2.3Materials required:

Micro organisms:

- *Escherichia coli(DH5 alpha)*
- *Bacillus subtilis* cultures isolated from soil

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Source of the organisms: *Tamil Nadu Agricultural University, Coimbatore (Tamil Nadu, India.)*

For the Extract:

Materials	***Quantity***
Allium sativum	1 kg
Catharanthus roseus	1kg
Ethanol (100%)	500mL
Pestle and Mortar	2 numbers
Sterile containers (with screw caps)	2numbers
Weighing machine	*1number*

For the Agar medium:

Materials	***Quantity***
Agar	80grams
Distilled water	5 litres
1000mL conical flask	4numbers
Yeast extract	12grams
Tryptone	20grams
NaCl	20grams

Others:

- Micropipettes – 100 µl (to spread the culture)

  10µl (to spot the extracts on the filter paper discs)
- Micropipette tips (one box of compatible tips for each pipette)
- Spread rod
- Alcohol a diffent alcohol ?
- Spirit lamps
- Filter paper discs
- Forceps ✓
- Disposable Gloves ✓

- Gentamicin sensitivity discs (readymade)
- Sterile inoculation discs
- Forceps
- Ruler
- Gloves

Appliances:

- Autoclave
- Incubator
- Laminar airflow chamber
- Refrigerator

No mention of autoclave ? Appendix A

2.4 Procedure[18]:

DAY1:

- The total number of Petri dishes that were required for the experiment were 200. The media required per plate is 20 mL. Therefore the total amount of media required was 4 litres.
- The media was prepared in four sets of apparatus simultaneously. A 1000mL conical flask was rinsed with distilled water and marked 'A'. 1000mL of distilled water was added to this flask after measuring using a measuring jar. To this, 5gms of Tryptone, 3gms of Yeast extract, 5gms of NaCl were added. The substances were measured using a weighing machine. Sterile tissue paper was used to weigh the substances that were then transferred into the conical flasks. 20gms of Agar was added to the flask. There were three other such 1000mL flasks that were taken and they were marked B, C, D and the same quantity of substances were taken.
- When the Agar was added, it was made sure that it did not stick to the sides of the flask. If it did, it was wiped using cotton. Cotton was rolled in such a way that it acted as a stopper for the flask. This was done to enable steam to pass through while autoclaving. Steam will help to kill any microbes present inside the flasks thereby creating a sterile environment. The Pestle and Mortar and the small glass tubes were enclosed within polythene covers. All of the apparatus i.e. the conical flasks (4+ A), the tubes (which will be used to store the extract) and the Pestle and Mortar were placed in the autoclave at 15lbs pressure (121°C). The outlet of the

[18] M.Dilikumar, Efficacy of leaf extracts of selected medicinal plants against multi-drug resistant strains of *Staphylococcus aureus*, Scientific transactions in Environment and Technovation, 2008, pp 23-27.

autoclave was closed after some steam escaped. After about half an hour, the apparatus was taken out of the autoclave and placed on a table. But, they were not removed from the plastic covers.

- Working in the sterile conditions of a laminar- airflow chamber, the solution from the flask A was poured into 50 sterile Petri dishes. This was done when the agar had cooled down slightly but was still in liquid form.

- These plates were left undisturbed for 15-20 minutes so as to allow it to set after which they were put in polythene covers and placed in the refrigerator at 4°C. The polythene covers help to prevent any moisture. Next, working in a laminar air-flow chamber (to keep the environment sterile), some bacteria were inoculated from the culture to a nutrient agar medium using sterile inoculation loops. The Petri dish was marked X and incubated at 37°C overnight (The temperature is ideal for the growth of Bacteria).

- The extracts were prepared using a Pestle and Mortar. The Garlic was chopped into tiny pieces (as they will be easy to grind). The *Catharanthus roseus* leaves were separated from their respective stems. 3gms of each substance was weighed and kept aside. 100% Ethanol (7mL) was added to each of the materials in the Pestle and Mortar so as to facilitate easier grinding. They were ground well and the extracts were transferred into small bottles and kept at 4°C overnight.

DAY 2:

- Working in a laminar air-flow chamber, respective bacteria were taken from the Petri dish X using an inoculation loop and introduced into the conical flasks labelled *E. coli* and *Bacillus*. It was made sure that a sufficient quantity of the bacteria culture was taken. The flask was shaken well.

- 200µl of the bacteria culture from the conical flasks labelled *Bacillus* and *E. coli* were added to the respective Petri dishes (Markings are made on the Petri dishes as shown in the figure 1 below) and spread evenly throughout using a spread rod and it was left to dry. The bottles with the extract were placed in a water bath of 65°C to evaporate ethanol. The spread rods were rinsed every time in ethanol (100%) before they were used to spread the bacteria.

- Discs were cut from filter papers using a punching machine. These discs were put in a small glass bottle and autoclaved.

- Meanwhile, the Petri dishes with the nutrient medium (prepared on day 1) were taken and 8 sections were marked on it using a permanent marker. For each of the extract, ten Petri dishes were used. The makings were done as shown below.

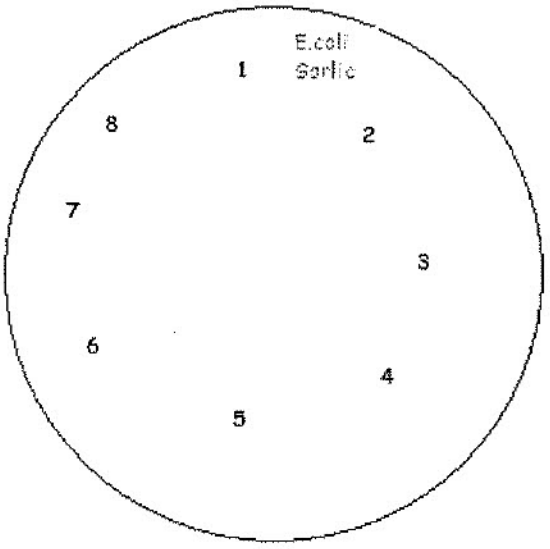

Figure 5 – Petri dish setup

The numerical represented the following:

1. Ethanol (control) ✓✓ —— on the dishes it is called Acetone (?) also in terms of data. Did you use acetone?
2. Gentamicin (antibiotic)
3. 10µl of the extract
4. 15µl of the extract

To study the antimicrobial action of every extract, 5 discs soaked in the same extract (say Garlic) were placed in every Petri dish at the marked positions. However, the amount of the extract taken was varied. The discs were soaked in the extracts with the help of a micropipette. There were 10 Petri dishes made for every extract so as to get 50 readings for the action of each extract on each bacterium. After the paper discs and the bacteria spread on the Petri dishes dried, the paper discs were placed on their respective numerals using forceps. Before doing so, the forceps were dipped in alcohol and exposed to the flame of a Bunsen burner every time so as to sterilize it. A Bunsen burner is used because the heat kills the cells of the bacteria. Alcohol is used because it is a surface sterilizing agent. After the discs were placed, ✓

the Petri dishes were placed in an incubator at 37°C. The results were observed the next day. The diameter of the halo around the antimicrobial extracts was measured using a ruler.

- Ethanol and disc were placed as control so as to ascertain that they themselves do not possess any antimicrobial properties. Gentamicin discs were placed to evaluate the efficacy of the plant extracts.

Discs with ethanol and Discs with nothing (dry) or what...?

2.5 Weakness and Improvement:

SNO	WEAKNESS	IMPROVEMENT
1	The extracts from plants are Crude extracts only.	The pure extract to be used for better results. Pure extracts can be prepared by using HPLC(High performance liquid Chromatography). HPLC is widely used in the fields of Biochemistry and analytical Chemistry to separate, identify and quantify compounds. This is a form of column chromatography. which comparative work you are ?
2	Stability of the result may vary depending upon the product composition, organisms and seasonal variations	The experiment can be performed throughout the year during different seasons and the results can be observed. The experiment may be carried against different pathogenic organisms isolated from different clinical specimens.

2.6 Statistical analysis:

Statistical analysis was performed using a two-tailed test

Mean[19] : $\frac{\sum x}{n}$

[19]Lipschutz,Seymour, Schiller,John Introduction to Probability and Statisctics, Tata McGraw-Hill Publishing company Ltd.New Delhi,2005,p5

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Standard deviation[20]: $$\frac{\sqrt{\sum(x1-\bar{x}1)^2+\sum(x2-\bar{x}2)^2}}{(n1+n2-2)}$$

$\bar{x}1$ = mean of the first sample

$\bar{x}2$ = mean of the second sample

n_1 = Size of the first sample [here n1 = 50]

n_2 = size of the second sample [here n2 = 50]

t-test: To test for the significance of the difference between two sample means, we use the following formula.

$$\frac{(mean1-mean2)\times\sqrt{n1*n2}}{s\times\sqrt{n1+n2}}$$

n1 = size of the first sample

n2= size of the second sample

s= standard deviation

Degree of freedom[21]

$Df = n_1 + n_2 - 2$

$= 98$

[20] Lipschutz,Seymour, Schiller,John Introduction to Probability and Statisctics, Tata McGraw-Hill Publishing company Ltd:New Delhi,2005,p9

[21] Damon,Andy,McGonegal,Randy,Tosto,Patricia,Biology developed specifically for the IB Diploma, Pearson Education Limited: UK,2007, pp1-10

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CHAPTER 3 : DATA COLLECTION AND PROCESSING:

Escherichia coli

Group Statistics	N	Mean	Std. Deviation
Gentamicin	50	22.1	0.81
Garlic on *E.coli* 10ul	50	21.24	1.15
Garlic on *E.coli* 15ul	50	23.98	1.41
Ethanol(control)	50	0 ·0 %	0.00

Gentamicin	50	23.78	0.97
Vinca rosea on *E.coli* 10ul	50	21.92	0.88
Vinca rosea on *E.coli* 15ul	50	23.72	0.88
Ethanol (control)	50	0	0.00

This is surprising since ethanol has antibacterial properties and can be used as a disinfectant.

Did the alcohol evaporate?

Bacillus subtilis

Group Statistics	N	Mean	Std. Deviation
Gentamicin	50	20.12	1.35
Garlic on *Bacillus* 10ul	50	21.64	1.14
Garlic on *Bacillus* 15ul	50	23.36	1.14
Ethanol(control)	50	0	0.00
Gentamicin	50	20.24	1.61
Vinca rosea on *Bacillus*10ul	50	21.46	1.66
Vinca rosea on *Bacillus*15ul	50	23.72	1.50
Ethanol (control)	50	0	0.00

Chapter 4: ANALYSIS AND INTERPRETATION

4.1 T-Value table

For *E.coli*

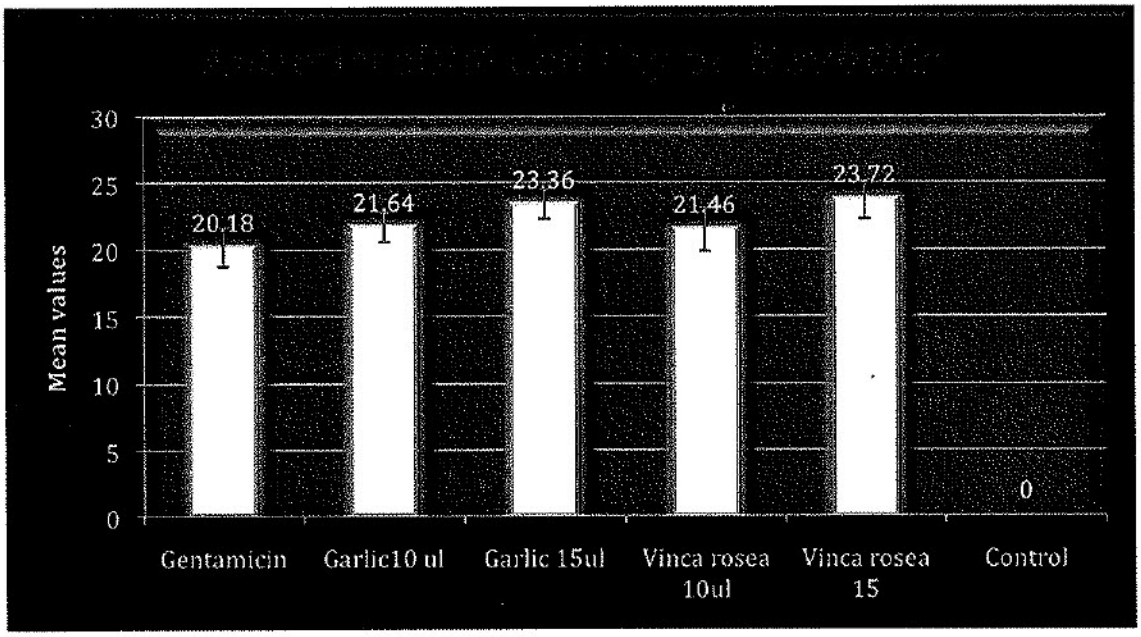
SNO	Comparison	t-value
1 a	Gentamicin and Garlic [10ul]	4.308
b	Gentamicin and Garlic [15ul]	8.178
2 a	Control and garlic [10ul]	130.281
b	Control and garlic [15ul]	120.529
3 a	Gentamicin and Vinca rosea[10ul]	10.030
b	Gentamicin and Vinca rosea [15ul]	0.323
4 a	control and Vinca rosea [10ul]	176.754
b	control and Vinca rosea [15ul]	190.261
5 a	Garlic and Vinca rosea [10ul]	3.320
b	Garlic and Vinca rosea [15ul]	1.107

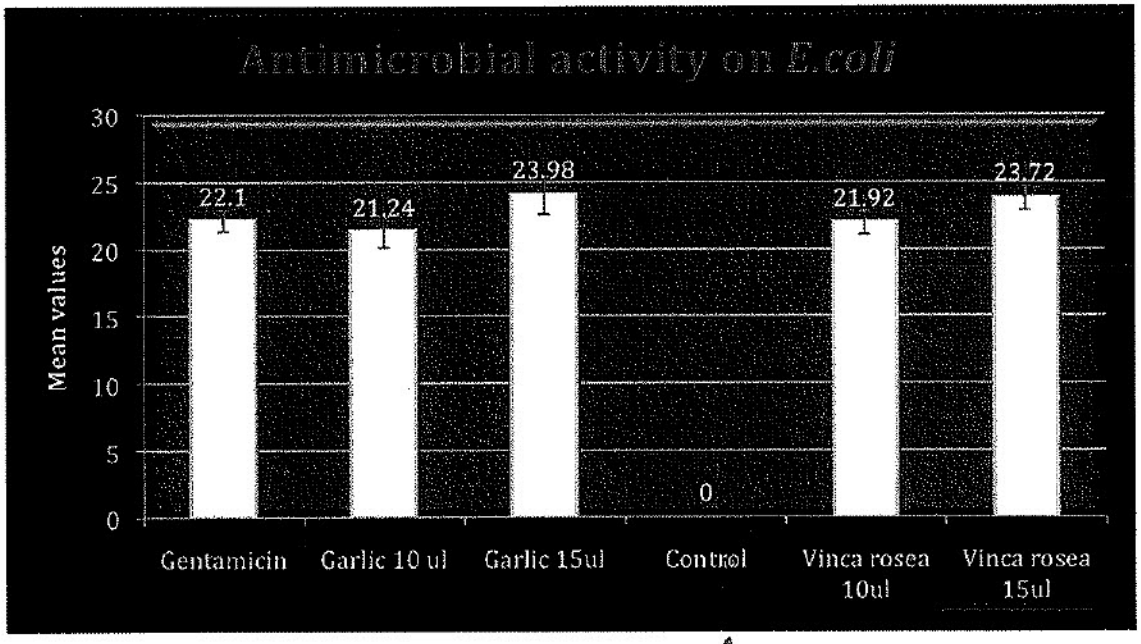
For *Bacillus*:

Sl.No.	Comparison	t-value
1. a.	Gentamicin and Garlic [10ul]	6.086
b.	Gentamicin and Garlic [15ul]	12.974
2. a.	Control and Garlic [10ul]	134.396
b.	Control and Garlic [15ul]	145.078
3. a.	Gentamicin and Vinca rosea [10ul]	3.734
b	Gentamicin and Vinca rosea [15ul]	11.184

4.a.	Control and Vinca rosea [10ul]	91.618
b	Control and Vinca rosea [15ul]	111.903
5. a.	Garlic and Vinca rosea [10ul]	0.633
b	Garlic and Vinca rosea [15ul]	1.352

4.2 GRAPHS:





must have evaporated?

4.3 Interpretation:

TESTING HYPOTHESIS 1:

Comparison of Control and Garlic:

Null hypothesis: The plant extracts will not have an effect on the growth of *Escherichia coli* and *Bacillus subtilis*

Positive hypothesis: The plant extracts will reduce the growth of *Escherichia coli* and *Bacillus subtilis*

On *E.coli*:

15µl:

In the comparison between Garlic and the control, the t-value obtained was 130.281 at the 98 degree of freedom [$p<0.05$] which is more than the table value. Therefore the positive hypothesis 1 can be accepted. The results indicate that Garlic reduces the growth of *E.coli*

10µl:

In the comparison between Garlic and the control, the t-value obtained was 120.529 at the 98 degree of freedom [$p<0.05$] which is more than the table value. Therefore the positive hypothesis1 can be accepted. The results indicate that Garlic reduces the growth of *E.coli.*

On *Bacillus*:

15µl:

In the comparison between the effect of Garlic and control, the t-values obtained were 134.396 at 98 degree of freedom[$p<0.05$] which is more than the table value. The positive hypothesis 1 can thus be accepted. The results indicate that Garlic reduces the growth of *B.subtilis*

10µl:

In the comparison between the effect of Garlic and control, the t-values obtained were 145.078 at 98 degree of freedom[$p<0.05$] which is more than the table value. The positive hypothesis1 can thus be accepted. The results indicate that Garlic reduces the growth of *B.subtilis*

Comparison between Control and Vinca rosea:

On *E.coli*:

15µl:

In the comparison between Vinca rosea and the control, the t values obtained was 176.754 at 98 degree of freedom[p<0.05] which is more than the table t-value. The positive hypothesis1 can therefore be accepted. The results indicate that Vinca rosea reduces the growth of *E.coli*

10µl:

In the comparison between Vinca rosea and the control, the t values obtained was and 190.261 at 98 degree of freedom [p<0.05] which are more than the table t-value. The positive hypothesis1 can therefore be accepted. The results indicate that Vinca rosea reduces the growth of *E.coli*

On *Bacillus*:

15µl:

In the comparison between the Control and Vinca rosea on *Bacillus*, the t-values obtained was 91.618 at 98 degrees of freedom [p<0.05] which is more than the table value and therefore the positive hypothesis1 can be accepted. The results indicate that Vinca rosea reduces the growth of B.subtilis

10µl:

In the comparison between the Control and Vinca rosea on *Bacillus*, the t-values obtained was 111.903 at 98 degrees of freedom [p<0.05] which is more than the table value and therefore the positive hypothesis1 can be accepted. The results indicate that Vinca rosea reduces the growth of B.subtilis

TESTING HYPOTHESIS 2:

Null hypothesis: There will be no difference in the antimicrobial activity of Gentamicin and the plant extract

Positive hypothesis: There will be a difference in the antimicrobial activity of Gentamicin and the plant extract

Comparison of Gentamicin and Garlic:

On *E.coli*:

15µl :

In the comparison between Gentamicin and Garlic the t-value obtained is 8.178 at the 98 degrees of freedom [p<0.05] which is less than the table value. Therefore, null hypothesis 2, which indicates that the effect of Garlic on *E.coli* is similar to the antibiotic, can be accepted.

10µl :

In the comparison between Gentamicin and Garlic the t-value obtained is 4.308 at the 98 degrees of freedom [p<0.05] which is less than the table value. Therefore, the null hypothesis 2, which indicates that the effect of Garlic on *E.coli* is similar to the antibiotic, can be accepted.

TESTING HYPOTHESIS 3:

Null Hypothesis: There is no difference in the antimicrobial effect of the two extracts.

Positive Hypothesis: There is a difference in the antimicrobial effect of the two extracts.

Comparison of Garlic and Vinca rosea:

On *E.coli*

15µl :

In the comparison of Garlic and Vinca rosea the t-value obtained is 3.320 at 98 degrees of freedom [$p<0.05$] which is less than the table value. Therefore the null hypothesis 3 can be accepted.

10µl:

In the comparison of Garlic and Vinca rosea the t-value obtained is 1.107 at 98 degrees of freedom [$p<0.05$] which is less than the table value. Therefore the null hypothesis 3 can be accepted.

On *Bacillus*

15µl:

In the comparison of Garlic and Vinca rosea the t-value obtained is 0.633 at 98 degree of freedom [$p<0.05$] which is less than the table value. Therefore the null hypothesis 3 can be accepted.

10µl:

In the comparison of Garlic and Vinca rosea the t-value obtained is 1.352 at 98 degree of freedom [$p<0.05$] which is less than the table value. Therefore the null hypothesis 3 can be accepted.

4.4 Discussion:

The results indicated that the plant extracts reduce the growth of *E.coli* and *Bacillus subtilis*. The effect was seen to increase with an increase in concentration of the two extracts. From the calculated t-values, it can be observed that the antimicrobial effect of the Garlic and Vinca rosea extracts are quite similar to that of Gentamicin (antibiotic). Plant extracts can thus be a natural alternative to antibiotics. The antimicrobial effect of Garlic is due to the presence of Allicin, which disrupts the cell membrane biosynthesis by interfering with the DNA and RNA synthesis[22]. From the bar graph, it can be noted that Garlic and Vinca rosea have a greater antimicrobial effect on *E.coli* (gram negative) than on *Bacillus subtilis* (gram positive). This result was similar to the result observed by Mr. Pankaj

Goyal[23]. It can therefore be concluded that *Bacillus* has a mild inhibitory effect[24]. This could be due to a difference in the cell wall composition between the two organisms[25]. The composition of the growth medium could potentially affect antimicrobial activity. As published in the Journal of ethnopharmacology, the antimicrobial effect of Garlic was found to be greater in media lacking tryptone or cysteine indicating that the effects might involve sulfhydryl reactivity[26]. Also, the efficacy of herbal compounds extracted from plants depends largely on the type of solvent used for extraction[27]. Here, the solvent used was 100% ethanol. Extracts prepared in methanol are known to have better antimicrobial activity. Inspite of me using a media that contained tryptone and preparing the extracts in 100% ethanol rather than in methanol, the antimicrobial effect observed was profound. The effect, however, could have been of a greater magnitude.

Chapter 5 - CONCLUSION:

The comparison of the 't' values indicated that the extracts of *Catharanthus roseus* and *Allium sativum* reduce the growth of *Escherichia coli* and *Bacillus subtilis* and that the effect is more with increase in concentration of the extracts. The plant extract show similar antimicrobial activity to the antibiotic Gentamicin. Also, both the plant extracts had a similar antimicrobial effect on both cultures. They both had a higher effect on *Escherichia coli* than *Bacillus subtilis*

5.1 Extensions:

- The efficacy of the plant extracts can be compared with antibiotics other than Gentamicin. The plant extract of Neem showed a very mild effect on the cultures of *E.coli* and *Bacillus* as observed in the pilot survey. However, the antimicrobial effect of *this extract* was not explored. Further studies on this could be done.
- The experiment's result can be observed by using 100% pure extracts. Ways to extract a 100% pure extract can be explored.

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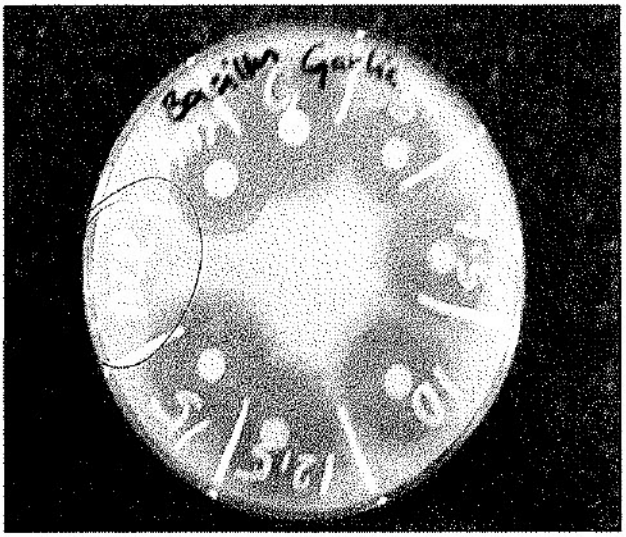

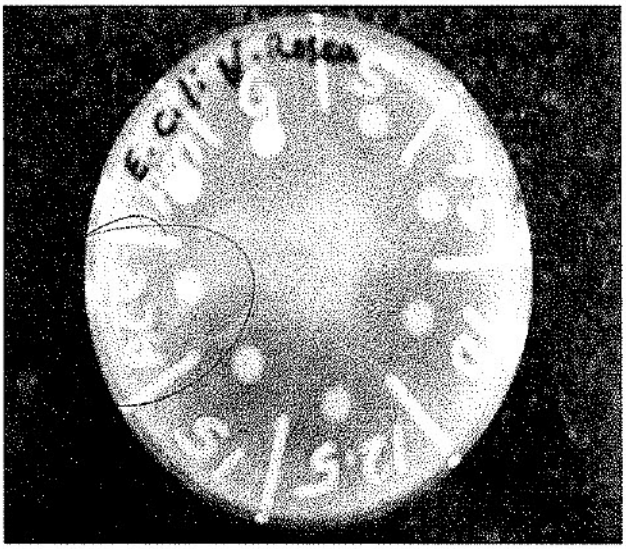

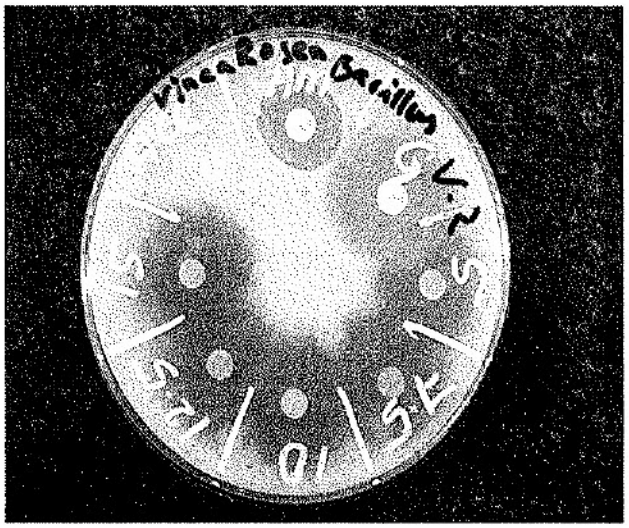

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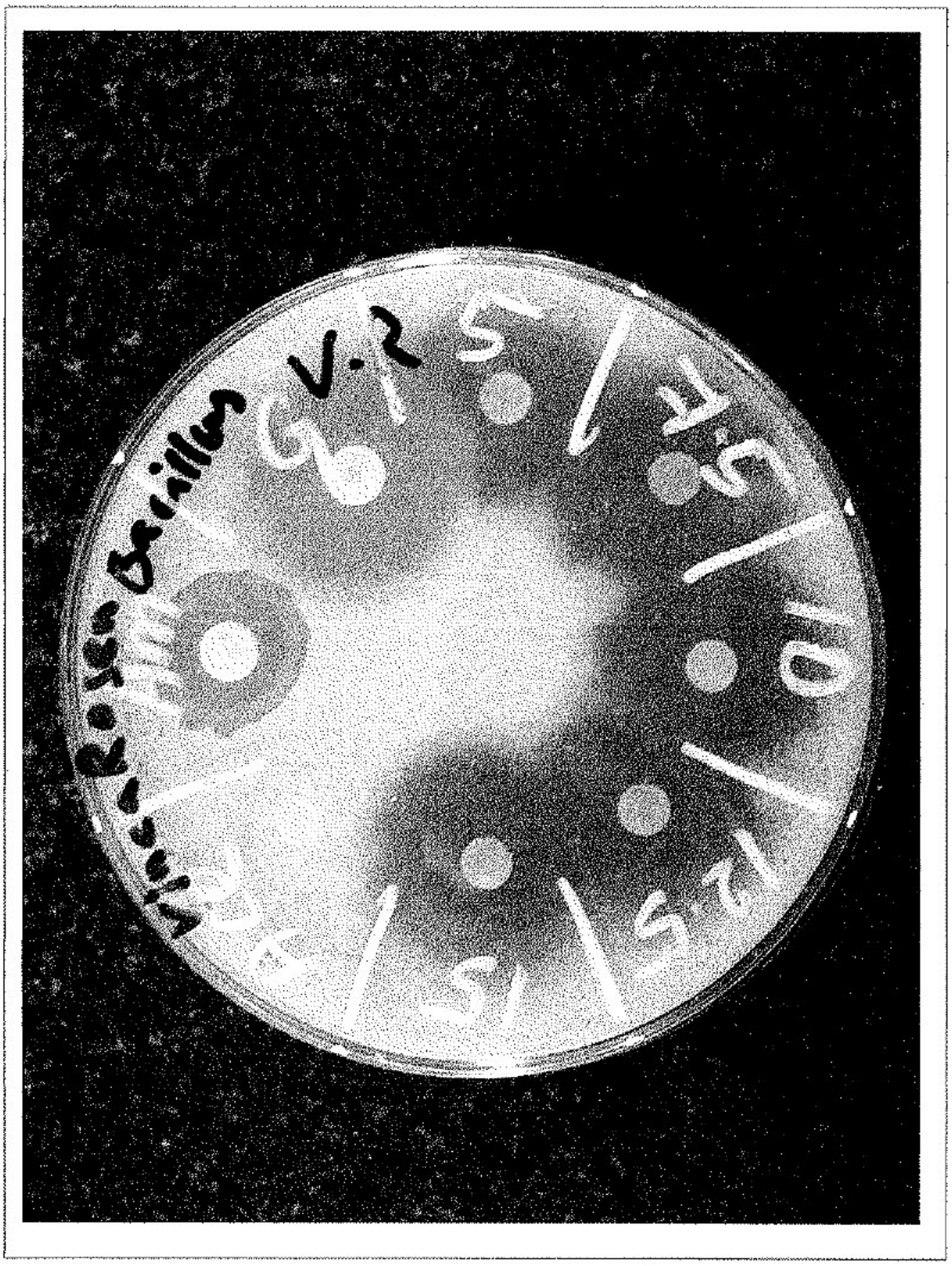
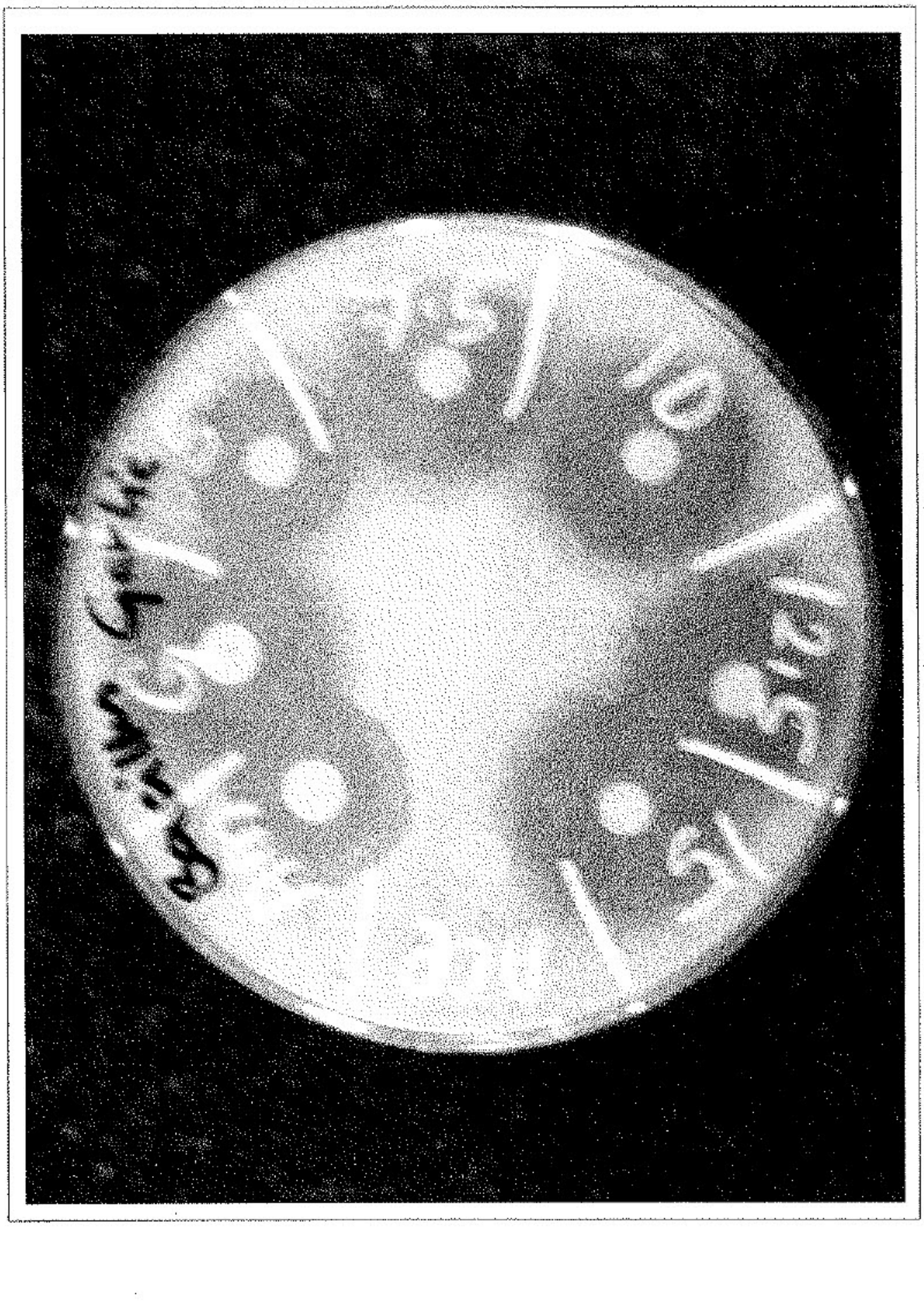
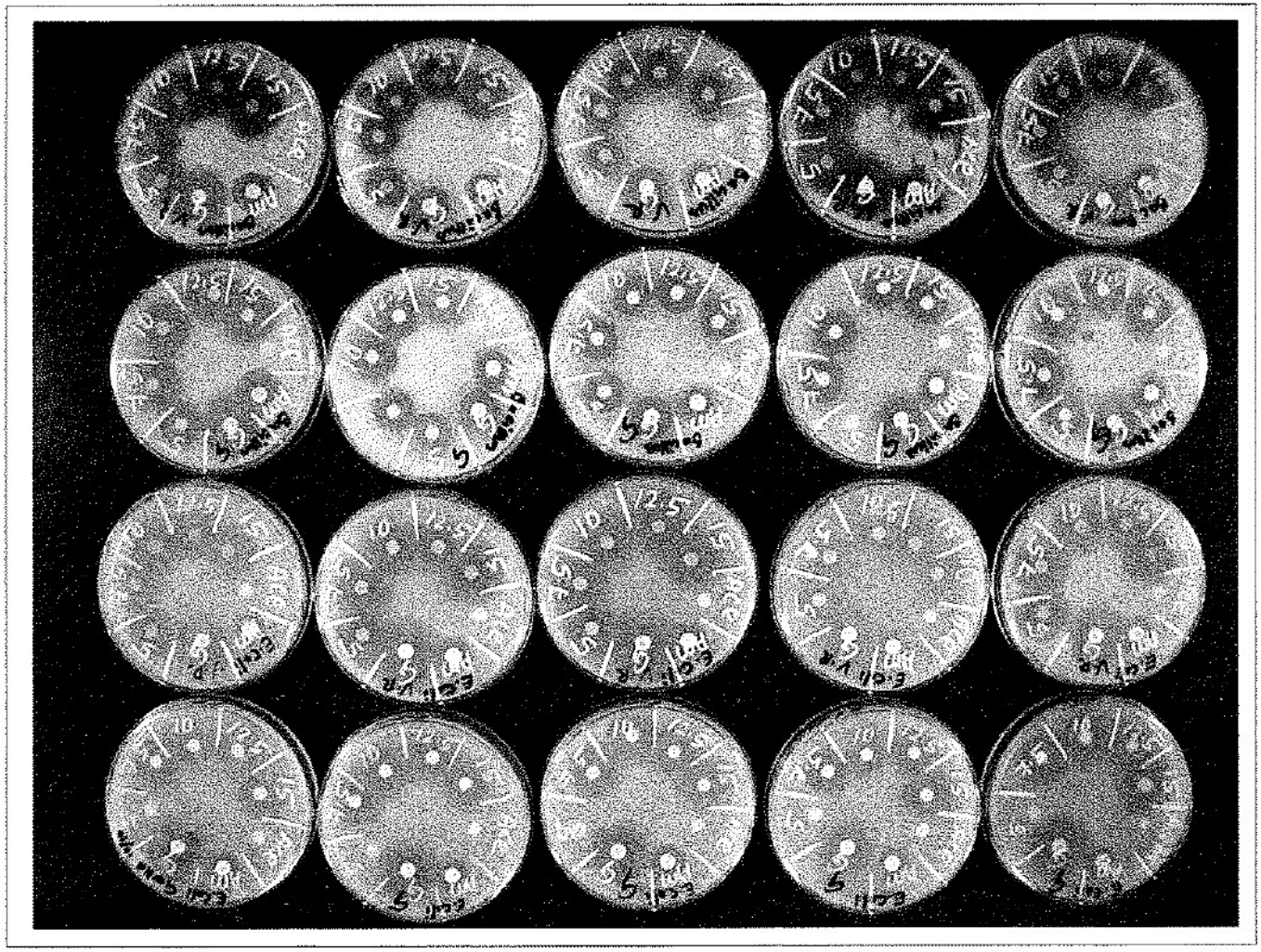

Vinca Rosea Bacillus V.2
5
7.5
10
12.5
15





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APPENDIX – A

Antimicrobial activity of Garlic on E.coli [n=50]								
A	Acetone	Ampicillin	Gentamyci	5ul	7.5ul	10 ul	12.5 ul	15 ul
Plate No. 1	0	15	23	0	21	22	25	26
Plate No. 2	0	14	22	0	22	22	22	23
Plate No. 3	0	14	22	0	20	20	22	23
Plate No. 4	0	14	22	0	22	23	23	23
Plate No. 5	0	15	21	0	17	17	19	20
Plate No. 6	0	15	23	0	20	21	21	23
Plate No. 7	0	15	22	0	21	21	21	23
Plate No. 8	0	16	23	0	20	23	23	25
Plate No. 9	0	15	23	0	21	22	23	24
Plate No. 10	0	15	23	0	20	21	22	23
Plate No. 11	0	16	21	0	20	20	22	24
Plate No. 12	0	15	22	0	21	21	22	23
Plate No. 13	0	14	21	0	21	21	23	23
Plate No. 14	0	15	23	0	20	21	21	24
Plate No. 15	0	15	23	0	19	22	23	26
Plate No. 16	0	15	23	0	20	21	22	24
Plate No. 17	0	15	23	0	21	23	23	25
Plate No. 18	0	16	22	0	18	20	21	25
Plate No. 19	0	14	21	0	19	20	23	24
Plate No. 20	0	15	22	0	18	21	22	24
Plate No. 21	0	15	22	0	20	21	23	24
Plate No. 22	0	16	22	0	21	21	22	23
Plate No. 23	0	14	21	0	20	21	21	24
Plate No. 24	0	15	21	0	18	22	22	25
Plate No. 25	0	14	23	0	19	20	20	23
Plate No. 26	0	15	23	0	18	23	23	24
Plate No. 27	0	15	21	0	18	23	23	25
Plate No. 28	0	15	22	0	21	22	23	26
Plate No. 29	0	16	22	0	22	23	23	24
Plate No. 30	0	15	23	0	20	21	21	22
Plate No. 31	0	15	23	0	18	20	20	22
Plate No. 32	0	14	21	0	20	21	21	21
Plate No. 33	0	14	22	0	19	20	24	25
Plate No. 34	0	15	23	0	18	21	23	26
Plate No. 35	0	14	22	0	18	21	24	26
Plate No. 36	0	15	23	0	21	23	23	25
Plate No. 37	0	16	22	0	18	22	23	26
Plate No. 38	0	15	22	0	21	22	22	23
Plate No. 39	0	14	22	0	21	23	23	24
Plate No. 40	0	14	23	0	22	22	23	23
Plate No. 41	0	15	21	0	22	22	23	26
Plate No. 42	0	15	21	0	21	21	22	24
Plate No. 43	0	16	23	0	19	21	22	26
Plate No. 44	0	14	23	0	21	21	21	23
Plate No. 45	0	14	23	0	20	20	21	23
Plate No. 46	0	14	21	0	20	20	24	26
Plate No. 47	0	14	21	0	21	21	22	23
Plate No. 48	0	15	22	0	20	21	21	23
Plate No. 49	0	14	21	0	20	20	22	23
Plate No. 50	0	16	21	0	20	21	23	26
Avg	0	15	22	0	20	21	22	24
SD	0	0.69	0.81	0.00	1.31	1.15	1.15	1.41

* All values are in mm

Antimicrobial activity of Garlic on Bacillus [n=50]

B	Acetone	Ampicillin	Gentamyci	5ul	7.5ul	10 ul	12.5 ul	15 ul
Plate No. 1	0	18	20	18	19	21	21	21
Plate No. 2	0	17	19	0	20	20	18	20
Plate No. 3	0	20	22	20	22	23	23	25
Plate No. 4	0	19	18	18	20	22	20	21
Plate No. 5	0	18	20	18	20	20	21	21
Plate No. 6	0	18	20	21	21	22	22	23
Plate No. 7	0	19	19	18	20	20	20	22
Plate No. 8	0	19	18	18	21	23	23	24
Plate No. 9	0	20	22	20	20	21	21	23
Plate No. 10	0	18	22	19	20	22	22	23
Plate No. 11	0	18	18	19	21	23	23	24
Plate No. 12	0	18	19	18	19	20	20	23
Plate No. 13	0	18	22	20	20	20	21	22
Plate No. 14	0	18	22	18	18	21	22	23
Plate No. 15	0	18	20	18	19	20	20	23
Plate No. 16	0	19	20	19	19	23	23	24
Plate No. 17	0	20	21	19	21	23	23	23
Plate No. 18	0	18	19	20	20	20	21	22
Plate No. 19	0	20	21	20	21	22	22	22
Plate No. 20	0	18	21	18	20	21	21	23
Plate No. 21	0	18	22	17	20	22	23	24
Plate No. 22	0	17	18	21	22	23	23	25
Plate No. 23	0	19	19	19	20	20	21	24
Plate No. 24	0	19	20	19	21	21	21	23
Plate No. 25	0	17	20	18	20	22	22	23
Plate No. 26	0	18	20	20	20	21	22	23
Plate No. 27	0	17	21	20	20	23	22	24
Plate No. 28	0	19	22	18	20	21	22	25
Plate No. 29	0	17	21	19	20	22	23	24
Plate No. 30	0	20	21	18	21	23	23	25
Plate No. 31	0	17	22	18	19	22	22	23
Plate No. 32	0	17	20	21	21	22	23	25
Plate No. 33	0	20	19	20	20	23	23	24
Plate No. 34	0	17	19	20	20	21	21	23
Plate No. 35	0	19	19	19	19	22	23	24
Plate No. 36	0	17	18	20	20	21	21	24
Plate No. 37	0	18	18	18	20	22	22	24
Plate No. 38	0	17	20	19	19	23	23	25
Plate No. 39	0	20	20	20	22	23	23	24
Plate No. 40	0	17	19	21	22	23	24	25
Plate No. 41	0	18	20	18	21	22	22	23
Plate No. 42	0	20	22	19	20	20	21	24
Plate No. 43	0	19	21	18	19	20	20	23
Plate No. 44	0	20	21	21	21	21	21	23
Plate No. 45	0	18	23	18	18	20	20	24
Plate No. 46	0	18	18	20	20	21	21	23
Plate No. 47	0	20	20	21	22	23	22	24
Plate No. 48	0	18	20	22	22	23	23	24
Plate No. 49	0	18	20	20	20	22	22	23
Plate No. 50	0	20	20	19	22	23	23	24
Avg	0	18	20	19	20	22	22	23
SD	0	1.07	1.35	2.95	1.02	1.14	1.20	1.14

* All values are in mm

Antimicrobial activity of Vinca rosea on E.coli [n=50]

C	Acetone	Ampicillin	Gentamyci	5ul	7.5ul	10 ul	12.5 ul	15 ul
Plate No. 1	0	15	26	20	22	22	23	25
Plate No. 2	0	14	24	19	20	20	21	21
Plate No. 3	0	14	24	20	20	22	25	25
Plate No. 4	0	15	23	20	20	22	24	24
Plate No. 5	0	15	24	20	20	23	23	24
Plate No. 6	0	15	24	19	20	21	22	23
Plate No. 7	0	14	25	20	20	20	22	24
Plate No. 8	0	15	24	20	20	21	21	23
Plate No. 9	0	15	26	20	20	22	22	24
Plate No. 10	0	14	24	20	20	22	23	25
Plate No. 11	0	15	24	19	20	21	22	23
Plate No. 12	0	15	26	18	19	20	23	24
Plate No. 13	0	14	24	20	20	22	22	24
Plate No. 14	0	14	23	19	21	22	22	23
Plate No. 15	0	15	23	20	21	23	23	23
Plate No. 16	0	14	23	20	21	23	23	24
Plate No. 17	0	15	24	19	20	22	22	22
Plate No. 18	0	15	23	20	20	23	23	23
Plate No. 19	0	14	23	20	20	22	24	24
Plate No. 20	0	15	24	19	21	22	23	23
Plate No. 21	0	14	25	20	20	22	23	24
Plate No. 22	0	14	23	20	21	23	24	25
Plate No. 23	0	15	23	20	20	22	22	23
Plate No. 24	0	14	25	19	21	23	23	24
Plate No. 25	0	15	23	20	20	22	24	24
Plate No. 26	0	15	23	19	20	21	22	23
Plate No. 27	0	15	24	20	20	22	22	23
Plate No. 28	0	14	23	19	20	22	23	24
Plate No. 29	0	14	23	20	21	23	23	24
Plate No. 30	0	15	25	20	21	23	24	25
Plate No. 31	0	14	23	19	20	22	24	26
Plate No. 32	0	14	23	19	20	23	24	24
Plate No. 33	0	15	23	21	20	22	24	23
Plate No. 34	0	15	24	20	21	23	24	24
Plate No. 35	0	15	23	19	21	23	23	23
Plate No. 36	0	15	23	20	20	21	22	24
Plate No. 37	0	14	24	19	22	22	23	24
Plate No. 38	0	14	25	20	21	21	23	23
Plate No. 39	0	15	23	20	22	22	23	23
Plate No. 40	0	15	26	19	21	22	23	23
Plate No. 41	0	15	23	20	22	23	23	24
Plate No. 42	0	14	23	20	21	21	22	23
Plate No. 43	0	14	24	20	20	20	23	24
Plate No. 44	0	15	25	20	21	22	24	24
Plate No. 45	0	14	23	20	20	22	24	25
Plate No. 46	0	15	24	19	21	21	23	23
Plate No. 47	0	15	23	20	20	22	24	24
Plate No. 48	0	14	24	20	20	21	23	23
Plate No. 49	0	15	23	19	21	22	23	24
Plate No. 50	0	15	22	18	20	23	24	25
Avg	0	15	24	20	20	22	23	24
SD	0	0.50	0.97	0.60	0.68	0.88	0.87	0.88

* All values are in mm

Antimicrobial activity of Vinca rosea on Bacillus [n=50]

D	Acetone	Ampicillin	Gentamyci	5ul	7.5ul	10 ul	12.5 ul	15 ul
Plate No. 1	0	17	18	17	17	18	20	21
Plate No. 2	0	17	17	17	17	20	17	20
Plate No. 3	0	17	18	17	17	18	18	18
Plate No. 4	0	17	24	23	24	25	27	28
Plate No. 5	0	17	24	20	24	25	25	25
Plate No. 6	0	17	17	18	19	20	20	23
Plate No. 7	0	17	18	19	19	19	20	22
Plate No. 8	0	16	18	18	19	19	20	23
Plate No. 9	0	16	19	19	20	20	20	22
Plate No. 10	0	17	21	20	20	20	21	23
Plate No. 11	0	16	23	21	21	23	23	24
Plate No. 12	0	17	22	21	21	23	24	24
Plate No. 13	0	16	21	22	23	24	25	25
Plate No. 14	0	17	20	21	22	23	23	24
Plate No. 15	0	16	21	20	21	23	23	24
Plate No. 16	0	17	20	21	21	22	23	23
Plate No. 17	0	16	22	21	21	22	23	23
Plate No. 18	0	16	23	18	20	21	22	24
Plate No. 19	0	16	21	17	19	20	22	23
Plate No. 20	0	16	21	18	19	20	22	23
Plate No. 21	0	16	22	18	19	20	23	24
Plate No. 22	0	17	21	18	20	21	23	24
Plate No. 23	0	17	21	17	19	20	22	23
Plate No. 24	0	16	21	17	20	22	23	23
Plate No. 25	0	16	23	18	20	23	23	23
Plate No. 26	0	16	21	17	19	22	23	24
Plate No. 27	0	16	20	18	18	23	24	24
Plate No. 28	0	16	20	17	19	22	24	24
Plate No. 29	0	16	20	18	20	20	22	23
Plate No. 30	0	16	20	17	20	22	24	25
Plate No. 31	0	17	18	18	20	21	23	25
Plate No. 32	0	16	19	18	20	23	24	25
Plate No. 33	0	16	18	18	20	22	23	25
Plate No. 34	0	17	20	17	19	21	23	25
Plate No. 35	0	17	18	17	17	20	24	25
Plate No. 36	0	17	20	18	20	21	23	25
Plate No. 37	0	16	20	18	20	22	25	25
Plate No. 38	0	17	20	19	19	21	24	25
Plate No. 39	0	17	20	21	21	23	23	24
Plate No. 40	0	16	21	17	20	21	24	25
Plate No. 41	0	16	20	20	20	20	22	23
Plate No. 42	0	17	20	23	23	23	23	24
Plate No. 43	0	16	20	23	23	23	23	24
Plate No. 44	0	16	20	22	24	24	25	25
Plate No. 45	0	16	20	22	23	23	23	24
Plate No. 46	0	17	20	23	23	23	25	25
Plate No. 47	0	17	20	17	20	20	21	23
Plate No. 48	0	17	20	19	20	20	23	23
Plate No. 49	0	17	21	20	20	21	21	24
Plate No. 50	0	16	20	20	21	21	22	23
Avg	0	16	20	19	20	21	23	24
SD	0	0.50	1.61	1.95	1.76	1.66	1.83	1.50

* All values are in mm

APPENDIX - B

SESSION : MAY 2009

A) Control vs Garlic on E.coli 10ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	23	22	1	1	0.81	0.58
	22	22	0	1	0.01	0.58
	22	20	0	-1	0.01	1.54
	22	23	0	2	0.01	3.10
	21	17	-1	-4	1.21	17.98
	23	21	1	0	0.81	0.06
	22	21	0	0	0.01	0.06
	23	23	1	2	0.81	3.10
	23	22	1	1	0.81	0.58
	23	21	1	0	0.81	0.06
	21	20	-1	-1	1.21	1.54
	22	21	0	0	0.01	0.06
	21	21	-1	0	1.21	0.06
	23	21	1	0	0.81	0.06
	23	22	1	1	0.81	0.58
	23	21	1	0	0.81	0.06
	23	23	1	2	0.81	3.10
	22	20	0	-1	0.01	1.54
	21	20	-1	-1	1.21	1.54
	22	21	0	0	0.01	0.06
	22	21	0	0	0.01	0.06
	22	21	0	0	0.01	0.06
	21	21	-1	0	1.21	0.06
	21	22	-1	1	1.21	0.58
	23	20	1	-1	0.81	1.54
	23	23	1	2	0.81	3.10
	21	23	-1	2	1.21	3.10
	22	22	0	1	0.01	0.58
	22	23	0	2	0.01	3.10
	23	21	1	0	0.81	0.06
	23	20	1	-1	0.81	1.54
	21	21	-1	0	1.21	0.06
	22	20	0	-1	0.01	1.54
	23	21	1	0	0.81	0.06
	22	21	0	0	0.01	0.06
	23	23	1	2	0.81	3.10
	22	22	0	1	0.01	0.58
	22	22	0	1	0.01	0.58
	22	23	0	2	0.01	3.10
	23	22	1	1	0.81	0.58
	21	22	-1	1	1.21	0.58
	21	21	-1	0	1.21	0.06
	23	21	1	0	0.81	0.06
	23	21	1	0	0.81	0.06
	23	20	1	-1	0.81	1.54
	21	20	-1	-1	1.21	1.54
	21	21	-1	0	1.21	0.06
	22	21	0	0	0.01	0.06
	21	20	-1	-1	1.21	1.54
	21	21	-1	0	1.21	0.06
Mean	22.1	21.24	0 total		32.5	65.12
SD	0.81	1.15				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 97.62

n1+n2-2 = 98

0.996122

S= 0.998059

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = 0.861672 0.86

sqrt of n1n2/n1+n2 = 25

5

t= 4.308361

B) Control vs Garlic on E.coli 15ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	23	26	1	2	0.81	4.08
	22	23	0	-1	0.01	0.96
	22	23	0	-1	0.01	0.96
	22	23	0	-1	0.01	0.96
	21	20	-1	-4	1.21	15.84
	23	23	1	-1	0.81	0.96
	22	23	0	-1	0.01	0.96
	23	25	1	1	0.81	1.04
	23	24	1	0	0.81	0.00
	23	23	1	-1	0.81	0.96
	21	24	-1	0	1.21	0.00
	22	23	0	-1	0.01	0.96
	21	23	-1	-1	1.21	0.96
	23	24	1	0	0.81	0.00
	23	26	1	2	0.81	4.08
	23	24	1	0	0.81	0.00
	23	25	1	1	0.81	1.04
	22	25	0	1	0.01	1.04
	21	24	-1	0	1.21	0.00
	22	24	0	0	0.01	0.00
	22	24	0	0	0.01	0.00
	22	23	0	-1	0.01	0.96
	21	24	-1	0	1.21	0.00
	21	25	-1	1	1.21	1.04
	23	23	1	-1	0.81	0.96
	23	24	1	0	0.81	0.00
	21	25	-1	1	1.21	1.04
	22	26	0	2	0.01	4.08
	22	24	0	0	0.01	0.00
	23	22	1	-2	0.81	3.92
	23	22	1	-2	0.81	3.92
	21	21	-1	-3	1.21	8.88
	22	25	0	1	0.01	1.04
	23	26	1	2	0.81	4.08
	22	26	0	2	0.01	4.08
	23	25	1	1	0.81	1.04
	22	26	0	2	0.01	4.08
	22	23	0	-1	0.01	0.96
	22	24	0	0	0.01	0.00
	23	23	1	-1	0.81	0.96
	21	26	-1	2	1.21	4.08
	21	24	-1	0	1.21	0.00
	23	26	1	2	0.81	4.08
	23	23	1	-1	0.81	0.96
	23	23	1	-1	0.81	0.96
	21	26	-1	2	1.21	4.08
	21	23	-1	-1	1.21	0.96
	22	23	0	-1	0.01	0.96
	21	23	-1	-1	1.21	0.96
	21	26	-1	2	1.21	4.08
Mean	22.1	23.98	0	total	32.5	96.98
SD	0.81	1.41				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 129.48

n1+n2-2 = 98

1.321224

S= 1.149445

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = -1.635572 0.86

sqrt of n1n2/n1+n2 = 25

5

t= -8.177858 t value be consider as positive

C)Neg control vs Garlic on E.coli 10ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	0	22	0	1	0	0.58
	0	22	0	1	0	0.58
	0	20	0	-1	0	1.54
	0	23	0	2	0	3.10
	0	17	0	-4	0	17.98
	0	21	0	0	0	0.06
	0	21	0	0	0	0.06
	0	23	0	2	0	3.10
	0	22	0	1	0	0.58
	0	21	0	0	0	0.06
	0	20	0	-1	0	1.54
	0	21	0	0	0	0.06
	0	21	0	0	0	0.06
	0	21	0	0	0	0.06
	0	22	0	1	0	0.58
	0	21	0	0	0	0.06
	0	23	0	2	0	3.10
	0	20	0	-1	0	1.54
	0	20	0	-1	0	1.54
	0	21	0	0	0	0.06
	0	21	0	0	0	0.06
	0	21	0	0	0	0.06
	0	21	0	0	0	0.06
	0	22	0	1	0	0.58
	0	20	0	-1	0	1.54
	0	23	0	2	0	3.10
	0	23	0	2	0	3.10
	0	22	0	1	0	0.58
	0	23	0	2	0	3.10
	0	21	0	0	0	0.06
	0	20	0	-1	0	1.54
	0	21	0	0	0	0.06
	0	20	0	-1	0	1.54
	0	21	0	0	0	0.06
	0	21	0	0	0	0.06
	0	23	0	2	0	3.10
	0	22	0	1	0	0.58
	0	22	0	1	0	0.58
	0	23	0	2	0	3.10
	0	22	0	1	0	0.58
	0	22	0	1	0	0.58
	0	21	0	0	0	0.06
	0	21	0	0	0	0.06
	0	21	0	0	0	0.06
	0	20	0	-1	0	1.54
	0	20	0	-1	0	1.54
	0	21	0	0	0	0.06
	0	21	0	0	0	0.06
	0	20	0	-1	0	1.54
	0	21	0	0	0	0.06
Mean	0.0	21.24	0	total	0	65.12
SD	0.00	1.15				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 65.12

n1+n2-2 = 98

0.66449

S= 0.815162

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = -26.05616 0.86

sqrt of n1n2/n1+n2 = 25

5

t= -130.2808

D) Neg control vs Garlic on E.coli 15ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	0	26	0	2	0	4.08
	0	23	0	-1	0	0.96
	0	23	0	-1	0	0.96
	0	23	0	-1	0	0.96
	0	20	0	-4	0	15.84
	0	23	0	-1	0	0.96
	0	23	0	-1	0	0.96
	0	25	0	1	0	1.04
	0	24	0	0	0	0.00
	0	23	0	-1	0	0.96
	0	24	0	0	0	0.00
	0	23	0	-1	0	0.96
	0	23	0	-1	0	0.96
	0	24	0	0	0	0.00
	0	26	0	2	0	4.08
	0	24	0	0	0	0.00
	0	25	0	1	0	1.04
	0	25	0	1	0	1.04
	0	24	0	0	0	0.00
	0	24	0	0	0	0.00
	0	24	0	0	0	0.00
	0	23	0	-1	0	0.96
	0	24	0	0	0	0.00
	0	25	0	1	0	1.04
	0	23	0	-1	0	0.96
	0	24	0	0	0	0.00
	0	25	0	1	0	1.04
	0	26	0	2	0	4.08
	0	24	0	0	0	0.00
	0	22	0	-2	0	3.92
	0	22	0	-2	0	3.92
	0	21	0	-3	0	8.88
	0	25	0	1	0	1.04
	0	26	0	2	0	4.08
	0	26	0	2	0	4.08
	0	25	0	1	0	1.04
	0	26	0	2	0	4.08
	0	23	0	-1	0	0.96
	0	24	0	0	0	0.00
	0	23	0	-1	0	0.96
	0	26	0	2	0	4.08
	0	24	0	0	0	0.00
	0	26	0	2	0	4.08
	0	23	0	-1	0	0.96
	0	23	0	-1	0	0.96
	0	26	0	2	0	4.08
	0	23	0	-1	0	0.96
	0	23	0	-1	0	0.96
	0	23	0	-1	0	0.96
	0	26	0	2	0	4.08
Mean	0.0	23.98	0	total	0	96.98
SD	0.00	1.41				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 96.98

n1+n2-2 = 98

0.989592

S= 0.994782

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = -24.10578 0.86

sqrt of n1n2/n1+n2 = 25

5

t= -120.5289 t value be consider as positive

E) Pos.control vsVinca rosea on E.coli 10ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	26	22	2	0	4.9284	0.01
	24	20	0	-2	0.0484	3.69
	24	22	0	0	0.0484	0.01
	23	22	-1	0	0.6084	0.01
	24	23	0	1	0.0484	1.17
	24	21	0	-1	0.0484	0.85
	25	20	1	-2	1.4884	3.69
	24	21	0	-1	0.0484	0.85
	26	22	2	0	4.9284	0.01
	24	22	0	0	0.0484	0.01
	24	21	0	-1	0.0484	0.85
	26	20	2	-2	4.9284	3.69
	24	22	0	0	0.0484	0.01
	23	22	-1	0	0.6084	0.01
	23	23	-1	1	0.6084	1.17
	23	23	-1	1	0.6084	1.17
	24	22	0	0	0.0484	0.01
	23	23	-1	1	0.6084	1.17
	23	22	-1	0	0.6084	0.01
	24	22	0	0	0.0484	0.01
	25	22	1	0	1.4884	0.01
	23	23	-1	1	0.6084	1.17
	23	22	-1	0	0.6084	0.01
	25	23	1	1	1.4884	1.17
	23	22	-1	0	0.6084	0.01
	23	21	-1	-1	0.6084	0.85
	24	22	0	0	0.0484	0.01
	23	22	-1	0	0.6084	0.01
	23	23	-1	1	0.6084	1.17
	25	23	1	1	1.4884	1.17
	23	22	-1	0	0.6084	0.01
	23	23	-1	1	0.6084	1.17
	23	22	-1	0	0.6084	0.01
	24	23	0	1	0.0484	1.17
	23	23	-1	1	0.6084	1.17
	23	21	-1	-1	0.6084	0.85
	24	22	0	0	0.0484	0.01
	25	21	1	-1	1.4884	0.85
	23	22	-1	0	0.6084	0.01
	26	22	2	0	4.9284	0.01
	23	23	-1	1	0.6084	1.17
	23	21	-1	-1	0.6084	0.85
	24	20	0	-2	0.0484	3.69
	25	22	1	0	1.4884	0.01
	23	22	-1	0	0.6084	0.01
	24	21	0	-1	0.0484	0.85
	23	22	-1	0	0.6084	0.01
	24	21	0	-1	0.0484	0.85
	23	22	-1	0	0.6084	0.01
	22	23	-2	1	3.1684	1.17
Mean	23.8	21.92	0 total		46.58	37.68
SD	0.97	0.88				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 84.26

n1+n2-2 = 98

0.859796

S= 0.927252

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = 2.005928 0.86

sqrt of n1n2/n1+n2 = 25

5

t= 10.02964

F) Pos.control vsVinca rosea on E.coli 15ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	26	25	2	1	4.9284	1.64
	24	21	0	-3	0.0484	7.40
	24	25	0	1	0.0484	1.64
	23	24	-1	0	0.6084	0.08
	24	24	0	0	0.0484	0.08
	24	23	0	-1	0.0484	0.52
	25	24	1	0	1.4884	0.08
	24	23	0	-1	0.0484	0.52
	26	24	2	0	4.9284	0.08
	24	25	0	1	0.0484	1.64
	24	23	0	-1	0.0484	0.52
	26	24	2	0	4.9284	0.08
	24	24	0	0	0.0484	0.08
	23	23	-1	-1	0.6084	0.52
	23	23	-1	-1	0.6084	0.52
	23	24	-1	0	0.6084	0.08
	24	22	0	-2	0.0484	2.96
	23	23	-1	-1	0.6084	0.52
	23	24	-1	0	0.6084	0.08
	24	23	0	-1	0.0484	0.52
	25	24	1	0	1.4884	0.08
	23	25	-1	1	0.6084	1.64
	23	23	-1	-1	0.6084	0.52
	25	24	1	0	1.4884	0.08
	23	24	-1	0	0.6084	0.08
	23	23	-1	-1	0.6084	0.52
	24	23	0	-1	0.0484	0.52
	23	24	-1	0	0.6084	0.08
	23	24	-1	0	0.6084	0.08
	25	25	1	1	1.4884	1.64
	23	26	-1	2	0.6084	5.20
	23	24	-1	0	0.6084	0.08
	23	23	-1	-1	0.6084	0.52
	24	24	0	0	0.0484	0.08
	23	23	-1	-1	0.6084	0.52
	23	24	-1	0	0.6084	0.08
	24	24	0	0	0.0484	0.08
	25	23	1	-1	1.4884	0.52
	23	23	-1	-1	0.6084	0.52
	26	23	2	-1	4.9284	0.52
	23	24	-1	0	0.6084	0.08
	23	23	-1	-1	0.6084	0.52
	24	24	0	0	0.0484	0.08
	25	24	1	0	1.4884	0.08
	23	25	-1	1	0.6084	1.64
	24	23	0	-1	0.0484	0.52
	23	24	-1	0	0.6084	0.08
	24	23	0	-1	0.0484	0.52
	23	24	-1	0	0.6084	0.08
	22	25	-2	1	3.1684	1.64
Mean	23.8	23.72	0	total	46.58	38.08
SD	0.97	0.88				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 84.66

n1+n2-2 = 98

0.863878

S= 0.92945

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = 0.064554 0.86

sqrt of n1n2/n1+n2 = 25

5

t= 0.322771

G) Neg.control vsVinca rosea on E.coli 10ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	0	22	0	0	0	0.01
	0	20	0	-2	0	3.69
	0	22	0	0	0	0.01
	0	22	0	0	0	0.01
	0	23	0	1	0	1.17
	0	21	0	-1	0	0.85
	0	20	0	-2	0	3.69
	0	21	0	-1	0	0.85
	0	22	0	0	0	0.01
	0	22	0	0	0	0.01
	0	21	0	-1	0	0.85
	0	20	0	-2	0	3.69
	0	22	0	0	0	0.01
	0	22	0	0	0	0.01
	0	23	0	1	0	1.17
	0	23	0	1	0	1.17
	0	22	0	0	0	0.01
	0	23	0	1	0	1.17
	0	22	0	0	0	0.01
	0	22	0	0	0	0.01
	0	22	0	0	0	0.01
	0	23	0	1	0	1.17
	0	22	0	0	0	0.01
	0	23	0	1	0	1.17
	0	22	0	0	0	0.01
	0	21	0	-1	0	0.85
	0	22	0	0	0	0.01
	0	22	0	0	0	0.01
	0	23	0	1	0	1.17
	0	23	0	1	0	1.17
	0	22	0	0	0	0.01
	0	23	0	1	0	1.17
	0	22	0	0	0	0.01
	0	23	0	1	0	1.17
	0	23	0	1	0	1.17
	0	21	0	-1	0	0.85
	0	22	0	0	0	0.01
	0	21	0	-1	0	0.85
	0	22	0	0	0	0.01
	0	22	0	0	0	0.01
	0	23	0	1	0	1.17
	0	21	0	-1	0	0.85
	0	20	0	-2	0	3.69
	0	22	0	0	0	0.01
	0	22	0	0	0	0.01
	0	21	0	-1	0	0.85
	0	22	0	0	0	0.01
	0	21	0	-1	0	0.85
	0	22	0	0	0	0.01
	0	23	0	1	0	1.17
Mean	0.0	21.92	0	total	0	37.68
SD	0.00	0.88				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 37.68

n1+n2-2 = 98

0.38449

S= 0.620072

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = -35.35071 0.86

sqrt of n1n2/n1+n2 = 25

5

t= -176.7535

H) Neg.control vsVinca rosea on E.coli 15ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	0	25	0	1	0	1.64
	0	21	0	-3	0	7.40
	0	25	0	1	0	1.64
	0	24	0	0	0	0.08
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	25	0	1	0	1.64
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	22	0	-2	0	2.96
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	25	0	1	0	1.64
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	24	0	0	0	0.08
	0	25	0	1	0	1.64
	0	26	0	2	0	5.20
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	23	0	-1	0	0.52
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	24	0	0	0	0.08
	0	25	0	1	0	1.64
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	25	0	1	0	1.64
Mean	0.0	23.72	0	total	0	38.08
SD	0.00	0.88				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 38.08

n1+n2-2 = 98

0.388571

S= 0.623355

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = -38.05215 0.86

sqrt of n1n2/n1+n2 = 25

5

t= -190.2608 t value be consider as positive

I) Garlic & vinca rosea 10ul on E.coli

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	22	22	1	0	0.5776	0.01
	22	20	1	-2	0.5776	3.69
	20	22	-1	0	1.5376	0.01
	23	22	2	0	3.0976	0.01
	17	23	-4	1	17.9776	1.17
	21	21	0	-1	0.0576	0.85
	21	20	0	-2	0.0576	3.69
	23	21	2	-1	3.0976	0.85
	22	22	1	0	0.5776	0.01
	21	22	0	0	0.0576	0.01
	20	21	-1	-1	1.5376	0.85
	21	20	0	-2	0.0576	3.69
	21	22	0	0	0.0576	0.01
	21	22	0	0	0.0576	0.01
	22	23	1	1	0.5776	1.17
	21	23	0	1	0.0576	1.17
	23	22	2	0	3.0976	0.01
	20	23	-1	1	1.5376	1.17
	20	22	-1	0	1.5376	0.01
	21	22	0	0	0.0576	0.01
	21	22	0	0	0.0576	0.01
	21	23	0	1	0.0576	1.17
	21	22	0	0	0.0576	0.01
	22	23	1	1	0.5776	1.17
	20	22	-1	0	1.5376	0.01
	23	21	2	-1	3.0976	0.85
	23	22	2	0	3.0976	0.01
	22	22	1	0	0.5776	0.01
	23	23	2	1	3.0976	1.17
	21	23	0	1	0.0576	1.17
	20	22	-1	0	1.5376	0.01
	21	23	0	1	0.0576	1.17
	20	22	-1	0	1.5376	0.01
	21	23	0	1	0.0576	1.17
	21	23	0	1	0.0576	1.17
	23	21	2	-1	3.0976	0.85
	22	22	1	0	0.5776	0.01
	22	21	1	-1	0.5776	0.85
	23	22	2	0	3.0976	0.01
	22	22	1	0	0.5776	0.01
	22	23	1	1	0.5776	1.17
	21	21	0	-1	0.0576	0.85
	21	20	0	-2	0.0576	3.69
	21	22	0	0	0.0576	0.01
	20	22	-1	0	1.5376	0.01
	20	21	-1	-1	1.5376	0.85
	21	22	0	0	0.0576	0.01
	21	21	0	-1	0.0576	0.85
	20	22	-1	0	1.5376	0.01
	21	23	0	1	0.0576	1.17
Mean	21.2	21.92	0	total	65.12	37.68
SD	1.15	0.88				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 102.80

n1+n2-2 = 98

1.04898

S= 1.024197

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = -0.663935 0.86

sqrt of n1n2/n1+n2 = 25

5

t= -3.319674

J) Garlic & vinca rosea 15ul on E.coli

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	26	25	2	1	4.0804	1.64
	23	21	-1	-3	0.9604	7.40
	23	25	-1	1	0.9604	1.64
	23	24	-1	0	0.9604	0.08
	20	24	-4	0	15.8404	0.08
	23	23	-1	-1	0.9604	0.52
	23	24	-1	0	0.9604	0.08
	25	23	1	-1	1.0404	0.52
	24	24	0	0	0.0004	0.08
	23	25	-1	1	0.9604	1.64
	24	23	0	-1	0.0004	0.52
	23	24	-1	0	0.9604	0.08
	23	24	-1	0	0.9604	0.08
	24	23	0	-1	0.0004	0.52
	26	23	2	-1	4.0804	0.52
	24	24	0	0	0.0004	0.08
	25	22	1	-2	1.0404	2.96
	25	23	1	-1	1.0404	0.52
	24	24	0	0	0.0004	0.08
	24	23	0	-1	0.0004	0.52
	24	24	0	0	0.0004	0.08
	23	25	-1	1	0.9604	1.64
	24	23	0	-1	0.0004	0.52
	25	24	1	0	1.0404	0.08
	23	24	-1	0	0.9604	0.08
	24	23	0	-1	0.0004	0.52
	25	23	1	-1	1.0404	0.52
	26	24	2	0	4.0804	0.08
	24	24	0	0	0.0004	0.08
	22	25	-2	1	3.9204	1.64
	22	26	-2	2	3.9204	5.20
	21	24	-3	0	8.8804	0.08
	25	23	1	-1	1.0404	0.52
	26	24	2	0	4.0804	0.08
	26	23	2	-1	4.0804	0.52
	25	24	1	0	1.0404	0.08
	26	24	2	0	4.0804	0.08
	23	23	-1	-1	0.9604	0.52
	24	23	0	-1	0.0004	0.52
	23	23	-1	-1	0.9604	0.52
	26	24	2	0	4.0804	0.08
	24	23	0	-1	0.0004	0.52
	26	24	2	0	4.0804	0.08
	23	24	-1	0	0.9604	0.08
	23	25	-1	1	0.9604	1.64
	26	23	2	-1	4.0804	0.52
	23	24	-1	0	0.9604	0.08
	23	23	-1	-1	0.9604	0.52
	23	24	-1	0	0.9604	0.08
	26	25	2	1	4.0804	1.64
Mean	24.0	23.72	0 total		96.98	38.08
SD	1.41	0.88				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 135.06

n1+n2-2 = 98

1.378163

S= 1.173952

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = 0.221474 0.86

sqrt of n1n2/n1+n2 = 25

5

t= 1.107371 t value be consider as positive

BACILLUS

K) Positive control vs Garlic on bacillus 10ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	20	21	0	-1	0.0144	0.41
	19	20	-1	-2	1.2544	2.69
	22	23	2	1	3.5344	1.85
	18	22	-2	0	4.4944	0.13
	20	20	0	-2	0.0144	2.69
	20	22	0	0	0.0144	0.13
	19	20	-1	-2	1.2544	2.69
	18	23	-2	1	4.4944	1.85
	22	21	2	-1	3.5344	0.41
	22	22	2	0	3.5344	0.13
	18	23	-2	1	4.4944	1.85
	19	20	-1	-2	1.2544	2.69
	22	20	2	-2	3.5344	2.69
	22	21	2	-1	3.5344	0.41
	20	20	0	-2	0.0144	2.69
	20	23	0	1	0.0144	1.85
	21	23	1	1	0.7744	1.85
	19	20	-1	-2	1.2544	2.69
	21	22	1	0	0.7744	0.13
	21	21	1	-1	0.7744	0.41
	22	22	2	0	3.5344	0.13
	18	23	-2	1	4.4944	1.85
	19	20	-1	-2	1.2544	2.69
	20	21	0	-1	0.0144	0.41
	20	22	0	0	0.0144	0.13
	20	21	0	-1	0.0144	0.41
	21	23	1	1	0.7744	1.85
	22	21	2	-1	3.5344	0.41
	21	22	1	0	0.7744	0.13
	21	23	1	1	0.7744	1.85
	22	22	2	0	3.5344	0.13
	20	22	0	0	0.0144	0.13
	19	23	-1	1	1.2544	1.85
	19	21	-1	-1	1.2544	0.41
	19	22	-1	0	1.2544	0.13
	18	21	-2	-1	4.4944	0.41
	18	22	-2	0	4.4944	0.13
	20	23	0	1	0.0144	1.85
	20	23	0	1	0.0144	1.85
	19	23	-1	1	1.2544	1.85
	20	22	0	0	0.0144	0.13
	22	20	2	-2	3.5344	2.69
	21	20	1	-2	0.7744	2.69
	21	21	1	-1	0.7744	0.41
	23	20	3	-2	8.2944	2.69
	18	21	-2	-1	4.4944	0.41
	20	23	0	1	0.0144	1.85
	20	23	0	1	0.0144	1.85
	20	22	0	0	0.0144	0.13
	20	23	0	1	0.0144	1.85
Mean	20.1	21.64	0	total	89.28	63.52
SD	1.35	1.14				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 152.80

n1+n2-2 = 98

1.559184

S= 1.248673

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = -1.217293 0.86

sqrt of n1n2/n1+n2 = 25

5

t= -6.086463

L) Positive control vs Garlic on bacillus 15ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	20	21	0	-2	0.0144	5.57
	19	20	-1	-3	1.2544	11.29
	22	25	2	2	3.5344	2.69
	18	21	-2	-2	4.4944	5.57
	20	21	0	-2	0.0144	5.57
	20	23	0	0	0.0144	0.13
	19	22	-1	-1	1.2544	1.85
	18	24	-2	1	4.4944	0.41
	22	23	2	0	3.5344	0.13
	22	23	2	0	3.5344	0.13
	18	24	-2	1	4.4944	0.41
	19	23	-1	0	1.2544	0.13
	22	22	2	-1	3.5344	1.85
	22	23	2	0	3.5344	0.13
	20	23	0	0	0.0144	0.13
	20	24	0	1	0.0144	0.41
	21	23	1	0	0.7744	0.13
	19	22	-1	-1	1.2544	1.85
	21	22	1	-1	0.7744	1.85
	21	23	1	0	0.7744	0.13
	22	24	2	1	3.5344	0.41
	18	25	-2	2	4.4944	2.69
	19	24	-1	1	1.2544	0.41
	20	23	0	0	0.0144	0.13
	20	23	0	0	0.0144	0.13
	20	23	0	0	0.0144	0.13
	21	24	1	1	0.7744	0.41
	22	25	2	2	3.5344	2.69
	21	24	1	1	0.7744	0.41
	21	25	1	2	0.7744	2.69
	22	23	2	0	3.5344	0.13
	20	25	0	2	0.0144	2.69
	19	24	-1	1	1.2544	0.41
	19	23	-1	0	1.2544	0.13
	19	24	-1	1	1.2544	0.41
	18	24	-2	1	4.4944	0.41
	18	24	-2	1	4.4944	0.41
	20	25	0	2	0.0144	2.69
	20	24	0	1	0.0144	0.41
	19	25	-1	2	1.2544	2.69
	20	23	0	0	0.0144	0.13
	22	24	2	1	3.5344	0.41
	21	23	1	0	0.7744	0.13
	21	23	1	0	0.7744	0.13
	23	24	3	1	8.2944	0.41
	18	23	-2	0	4.4944	0.13
	20	24	0	1	0.0144	0.41
	20	24	0	1	0.0144	0.41
	20	23	0	0	0.0144	0.13
	20	24	0	1	0.0144	0.41
Mean	20.1	23.36	0	total	89.28	63.52
SD	1.35	1.14				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 152.80

n1+n2-2 = 98

1.559184

S= 1.248673

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = -2.594755 0.86

sqrt of n1n2/n1+n2 = 25

5

t= -12.97378 t value be consider as positive

M) Neg control vs Garlic on bacillus 10ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	0	21	0	-1	0	0.41
	0	20	0	-2	0	2.69
	0	23	0	1	0	1.85
	0	22	0	0	0	0.13
	0	20	0	-2	0	2.69
	0	22	0	0	0	0.13
	0	20	0	-2	0	2.69
	0	23	0	1	0	1.85
	0	21	0	-1	0	0.41
	0	22	0	0	0	0.13
	0	23	0	1	0	1.85
	0	20	0	-2	0	2.69
	0	20	0	-2	0	2.69
	0	21	0	-1	0	0.41
	0	20	0	-2	0	2.69
	0	23	0	1	0	1.85
	0	23	0	1	0	1.85
	0	20	0	-2	0	2.69
	0	22	0	0	0	0.13
	0	21	0	-1	0	0.41
	0	22	0	0	0	0.13
	0	23	0	1	0	1.85
	0	20	0	-2	0	2.69
	0	21	0	-1	0	0.41
	0	22	0	0	0	0.13
	0	21	0	-1	0	0.41
	0	23	0	1	0	1.85
	0	21	0	-1	0	0.41
	0	22	0	0	0	0.13
	0	23	0	1	0	1.85
	0	22	0	0	0	0.13
	0	22	0	0	0	0.13
	0	23	0	1	0	1.85
	0	21	0	-1	0	0.41
	0	22	0	0	0	0.13
	0	21	0	-1	0	0.41
	0	22	0	0	0	0.13
	0	23	0	1	0	1.85
	0	23	0	1	0	1.85
	0	23	0	1	0	1.85
	0	22	0	0	0	0.13
	0	20	0	-2	0	2.69
	0	20	0	-2	0	2.69
	0	21	0	-1	0	0.41
	0	20	0	-2	0	2.69
	0	21	0	-1	0	0.41
	0	23	0	1	0	1.85
	0	23	0	1	0	1.85
	0	22	0	0	0	0.13
	0	23	0	1	0	1.85
Mean	0.0	21.64	0	total	0	63.52
SD	0.00	1.14				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 63.52

n1+n2-2 = 98

0.648163

S= 0.805086

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = -26.87912 0.86

sqrt of n1n2/n1+n2 = 25

5

t= -134.3956

N) Neg control vs Garlic on bacillus 15ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	0	21	0	-2	0	5.57
	0	20	0	-3	0	11.29
	0	25	0	2	0	2.69
	0	21	0	-2	0	5.57
	0	21	0	-2	0	5.57
	0	23	0	0	0	0.13
	0	22	0	-1	0	1.85
	0	24	0	1	0	0.41
	0	23	0	0	0	0.13
	0	23	0	0	0	0.13
	0	24	0	1	0	0.41
	0	23	0	0	0	0.13
	0	22	0	-1	0	1.85
	0	23	0	0	0	0.13
	0	23	0	0	0	0.13
	0	24	0	1	0	0.41
	0	23	0	0	0	0.13
	0	22	0	-1	0	1.85
	0	22	0	-1	0	1.85
	0	23	0	0	0	0.13
	0	24	0	1	0	0.41
	0	25	0	2	0	2.69
	0	24	0	1	0	0.41
	0	23	0	0	0	0.13
	0	23	0	0	0	0.13
	0	23	0	0	0	0.13
	0	24	0	1	0	0.41
	0	25	0	2	0	2.69
	0	24	0	1	0	0.41
	0	25	0	2	0	2.69
	0	23	0	0	0	0.13
	0	25	0	2	0	2.69
	0	24	0	1	0	0.41
	0	23	0	0	0	0.13
	0	24	0	1	0	0.41
	0	24	0	1	0	0.41
	0	24	0	1	0	0.41
	0	25	0	2	0	2.69
	0	24	0	1	0	0.41
	0	25	0	2	0	2.69
	0	23	0	0	0	0.13
	0	24	0	1	0	0.41
	0	23	0	0	0	0.13
	0	23	0	0	0	0.13
	0	24	0	1	0	0.41
	0	23	0	0	0	0.13
	0	24	0	1	0	0.41
	0	24	0	1	0	0.41
	0	23	0	0	0	0.13
	0	24	0	1	0	0.41
Mean	0.0	23.36	0 total		0	63.52
SD	0.00	1.14				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 63.52

n1+n2-2 = 98

0.648163

S= 0.805086

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = -29.01554 0.86

sqrt of n1n2/n1+n2 = 25

5

t= -145.0777 t value be consider as positive

O) Pos.control vsVinca rosea on Bacillus 10ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	18	18	-2	-3	5.0176	11.97
	17	20	-3	-1	10.4976	2.13
	18	18	-2	-3	5.0176	11.97
	24	25	4	4	14.1376	12.53
	24	25	4	4	14.1376	12.53
	17	20	-3	-1	10.4976	2.13
	18	19	-2	-2	5.0176	6.05
	18	19	-2	-2	5.0176	6.05
	19	20	-1	-1	1.5376	2.13
	21	20	1	-1	0.5776	2.13
	23	23	3	2	7.6176	2.37
	22	23	2	2	3.0976	2.37
	21	24	1	3	0.5776	6.45
	20	23	0	2	0.0576	2.37
	21	23	1	2	0.5776	2.37
	20	22	0	1	0.0576	0.29
	22	22	2	1	3.0976	0.29
	23	21	3	0	7.6176	0.21
	21	20	1	-1	0.5776	2.13
	21	20	1	-1	0.5776	2.13
	22	20	2	-1	3.0976	2.13
	21	21	1	0	0.5776	0.21
	21	20	1	-1	0.5776	2.13
	21	22	1	1	0.5776	0.29
	23	23	3	2	7.6176	2.37
	21	22	1	1	0.5776	0.29
	20	23	0	2	0.0576	2.37
	20	22	0	1	0.0576	0.29
	20	20	0	-1	0.0576	2.13
	20	22	0	1	0.0576	0.29
	18	21	-2	0	5.0176	0.21
	19	23	-1	2	1.5376	2.37
	18	22	-2	1	5.0176	0.29
	20	21	0	0	0.0576	0.21
	18	20	-2	-1	5.0176	2.13
	20	21	0	0	0.0576	0.21
	20	22	0	1	0.0576	0.29
	20	21	0	0	0.0576	0.21
	20	23	0	2	0.0576	2.37
	21	21	1	0	0.5776	0.21
	20	20	0	-1	0.0576	2.13
	20	23	0	2	0.0576	2.37
	20	23	0	2	0.0576	2.37
	20	24	0	3	0.0576	6.45
	20	23	0	2	0.0576	2.37
	20	23	0	2	0.0576	2.37
	20	20	0	-1	0.0576	2.13
	20	20	0	-1	0.0576	2.13
	21	21	1	0	0.5776	0.21
	20	21	0	0	0.0576	0.21
Mean	20.2	21.46	0 total		127.12	134.42
SD	1.61	1.66				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 261.54

n1+n2-2 = 98

2.668776

S= 1.633639

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = -0.746799 0.86

sqrt of n1n2/n1+n2 = 25

5

t= -3.733996

P) Pos.control vsVinca rosea on Bacillus 15ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	18	21	-2	-3	5.0176	7.40
	17	20	-3	-4	10.4976	13.84
	18	18	-2	-6	5.0176	32.72
	24	28	4	4	14.1376	18.32
	24	25	4	1	14.1376	1.64
	17	23	-3	-1	10.4976	0.52
	18	22	-2	-2	5.0176	2.96
	18	23	-2	-1	5.0176	0.52
	19	22	-1	-2	1.5376	2.96
	21	23	1	-1	0.5776	0.52
	23	24	3	0	7.6176	0.08
	22	24	2	0	3.0976	0.08
	21	25	1	1	0.5776	1.64
	20	24	0	0	0.0576	0.08
	21	24	1	0	0.5776	0.08
	20	23	0	-1	0.0576	0.52
	22	23	2	-1	3.0976	0.52
	23	24	3	0	7.6176	0.08
	21	23	1	-1	0.5776	0.52
	21	23	1	-1	0.5776	0.52
	22	24	2	0	3.0976	0.08
	21	24	1	0	0.5776	0.08
	21	23	1	-1	0.5776	0.52
	21	23	1	-1	0.5776	0.52
	23	23	3	-1	7.6176	0.52
	21	24	1	0	0.5776	0.08
	20	24	0	0	0.0576	0.08
	20	24	0	0	0.0576	0.08
	20	23	0	-1	0.0576	0.52
	20	25	0	1	0.0576	1.64
	18	25	-2	1	5.0176	1.64
	19	25	-1	1	1.5376	1.64
	18	25	-2	1	5.0176	1.64
	20	25	0	1	0.0576	1.64
	18	25	-2	1	5.0176	1.64
	20	25	0	1	0.0576	1.64
	20	25	0	1	0.0576	1.64
	20	25	0	1	0.0576	1.64
	20	24	0	0	0.0576	0.08
	21	25	1	1	0.5776	1.64
	20	23	0	-1	0.0576	0.52
	20	24	0	0	0.0576	0.08
	20	24	0	0	0.0576	0.08
	20	25	0	1	0.0576	1.64
	20	24	0	0	0.0576	0.08
	20	25	0	1	0.0576	1.64
	20	23	0	-1	0.0576	0.52
	20	23	0	-1	0.0576	0.52
	21	24	1	0	0.5776	0.08
	20	23	0	-1	0.0576	0.52
Mean	20.2	23.72	0	total	127.12	110.08
SD	1.61	1.50				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 237.20

n1+n2-2 = 98

2.420408

S= 1.555766

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = -2.23684 0.86

sqrt of n1n2/n1+n2 = 25

5

t= -11.1842 t value be consider as positive

Q) Neg.control vsVinca rosea on Bacillus 10ul						
	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	0	18	0	-3	0	11.97
	0	20	0	-1	0	2.13
	0	18	0	-3	0	11.97
	0	25	0	4	0	12.53
	0	25	0	4	0	12.53
	0	20	0	-1	0	2.13
	0	19	0	-2	0	6.05
	0	19	0	-2	0	6.05
	0	20	0	-1	0	2.13
	0	20	0	-1	0	2.13
	0	23	0	2	0	2.37
	0	23	0	2	0	2.37
	0	24	0	3	0	6.45
	0	23	0	2	0	2.37
	0	23	0	2	0	2.37
	0	22	0	1	0	0.29
	0	22	0	1	0	0.29
	0	21	0	0	0	0.21
	0	20	0	-1	0	2.13
	0	20	0	-1	0	2.13
	0	20	0	-1	0	2.13
	0	21	0	0	0	0.21
	0	20	0	-1	0	2.13
	0	22	0	1	0	0.29
	0	23	0	2	0	2.37
	0	22	0	1	0	0.29
	0	23	0	2	0	2.37
	0	22	0	1	0	0.29
	0	20	0	-1	0	2.13
	0	22	0	1	0	0.29
	0	21	0	0	0	0.21
	0	23	0	2	0	2.37
	0	22	0	1	0	0.29
	0	21	0	0	0	0.21
	0	20	0	-1	0	2.13
	0	21	0	0	0	0.21
	0	22	0	1	0	0.29
	0	21	0	0	0	0.21
	0	23	0	2	0	2.37
	0	21	0	0	0	0.21
	0	20	0	-1	0	2.13
	0	23	0	2	0	2.37
	0	23	0	2	0	2.37
	0	24	0	3	0	6.45
	0	23	0	2	0	2.37
	0	23	0	2	0	2.37
	0	20	0	-1	0	2.13
	0	20	0	-1	0	2.13
	0	21	0	0	0	0.21
	0	21	0	0	0	0.21
Mean	0.0	21.46	0 total		0	134.42
SD	0.00	1.66				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) =		134.42
n1+n2-2 =		98
		1.371633
S=		1.171167

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S =	-18.3236	0.86
sqrt of n1n2/n1+n2 =	25	
	5	
t=	-91.618	

R) Neg.control vsVinca rosea on Bacillus 15ul						
	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	0	21	0	-3	0	7.40
	0	20	0	-4	0	13.84
	0	18	0	-6	0	32.72
	0	28	0	4	0	18.32
	0	25	0	1	0	1.64
	0	23	0	-1	0	0.52
	0	22	0	-2	0	2.96
	0	23	0	-1	0	0.52
	0	22	0	-2	0	2.96
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	24	0	0	0	0.08
	0	25	0	1	0	1.64
	0	24	0	0	0	0.08
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	23	0	-1	0	0.52
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	24	0	0	0	0.08
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
	0	25	0	1	0	1.64
	0	25	0	1	0	1.64
	0	25	0	1	0	1.64
	0	25	0	1	0	1.64
	0	25	0	1	0	1.64
	0	25	0	1	0	1.64
	0	25	0	1	0	1.64
	0	25	0	1	0	1.64
	0	25	0	1	0	1.64
	0	24	0	0	0	0.08
	0	25	0	1	0	1.64
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	24	0	0	0	0.08
	0	25	0	1	0	1.64
	0	24	0	0	0	0.08
	0	25	0	1	0	1.64
	0	23	0	-1	0	0.52
	0	23	0	-1	0	0.52
	0	24	0	0	0	0.08
	0	23	0	-1	0	0.52
Mean	0.0	23.72	0 total		0	110.08
SD	0.00	1.50				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) =		110.08
n1+n2-2 =		98
		1.123265
S=		1.059842

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S =	-22.38069	0.86
sqrt of n1n2/n1+n2 =	25	
	5	
t=	-111.9035	t value be consider as positive

S) Garlic & vinca rosea 10ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	21	18	-1	-3	0.4096	11.97
	20	20	-2	-1	2.6896	2.13
	23	18	1	-3	1.8496	11.97
	22	25	0	4	0.1296	12.53
	20	25	-2	4	2.6896	12.53
	22	20	0	-1	0.1296	2.13
	20	19	-2	-2	2.6896	6.05
	23	19	1	-2	1.8496	6.05
	21	20	-1	-1	0.4096	2.13
	22	20	0	-1	0.1296	2.13
	23	23	1	2	1.8496	2.37
	20	23	-2	2	2.6896	2.37
	20	24	-2	3	2.6896	6.45
	21	23	-1	2	0.4096	2.37
	20	23	-2	2	2.6896	2.37
	23	22	1	1	1.8496	0.29
	23	22	1	1	1.8496	0.29
	20	21	-2	0	2.6896	0.21
	22	20	0	-1	0.1296	2.13
	21	20	-1	-1	0.4096	2.13
	22	20	0	-1	0.1296	2.13
	23	21	1	0	1.8496	0.21
	20	20	-2	-1	2.6896	2.13
	21	22	-1	1	0.4096	0.29
	22	23	0	2	0.1296	2.37
	21	22	-1	1	0.4096	0.29
	23	23	1	2	1.8496	2.37
	21	22	-1	1	0.4096	0.29
	22	20	0	-1	0.1296	2.13
	23	22	1	1	1.8496	0.29
	22	21	0	0	0.1296	0.21
	22	23	0	2	0.1296	2.37
	23	22	1	1	1.8496	0.29
	21	21	-1	0	0.4096	0.21
	22	20	0	-1	0.1296	2.13
	21	21	-1	0	0.4096	0.21
	22	22	0	1	0.1296	0.29
	23	21	1	0	1.8496	0.21
	23	23	1	2	1.8496	2.37
	23	21	1	0	1.8496	0.21
	22	20	0	-1	0.1296	2.13
	20	23	-2	2	2.6896	2.37
	20	23	-2	2	2.6896	2.37
	21	24	-1	3	0.4096	6.45
	20	23	-2	2	2.6896	2.37
	21	23	-1	2	0.4096	2.37
	23	20	1	-1	1.8496	2.13
	23	20	1	-1	1.8496	2.13
	22	21	0	0	0.1296	0.21
	23	21	1	0	1.8496	0.21
Mean	21.6	21.46	0	total	63.52	134.42
SD	1.14	1.66				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 197.94

n1+n2-2 = 98

2.019796

S= 1.421195

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = 0.126654 0.86

sqrt of n1n2/n1+n2 = 25

5

t= 0.63327

T) Garlic & vinca rosea 15ul

	x1	x2	d1=x1-X1	d2=x2-X2	d1xd1	d2xd2
	21	21	-2	-3	5.5696	7.40
	20	20	-3	-4	11.2896	13.84
	25	18	2	-6	2.6896	32.72
	21	28	-2	4	5.5696	18.32
	21	25	-2	1	5.5696	1.64
	23	23	0	-1	0.1296	0.52
	22	22	-1	-2	1.8496	2.96
	24	23	1	-1	0.4096	0.52
	23	22	0	-2	0.1296	2.96
	23	23	0	-1	0.1296	0.52
	24	24	1	0	0.4096	0.08
	23	24	0	0	0.1296	0.08
	22	25	-1	1	1.8496	1.64
	23	24	0	0	0.1296	0.08
	23	24	0	0	0.1296	0.08
	24	23	1	-1	0.4096	0.52
	23	23	0	-1	0.1296	0.52
	22	24	-1	0	1.8496	0.08
	22	23	-1	-1	1.8496	0.52
	23	23	0	-1	0.1296	0.52
	24	24	1	0	0.4096	0.08
	25	24	2	0	2.6896	0.08
	24	23	1	-1	0.4096	0.52
	23	23	0	-1	0.1296	0.52
	23	23	0	-1	0.1296	0.52
	23	24	0	0	0.1296	0.08
	24	24	1	0	0.4096	0.08
	25	24	2	0	2.6896	0.08
	24	23	1	-1	0.4096	0.52
	25	25	2	1	2.6896	1.64
	23	25	0	1	0.1296	1.64
	25	25	2	1	2.6896	1.64
	24	25	1	1	0.4096	1.64
	23	25	0	1	0.1296	1.64
	24	25	1	1	0.4096	1.64
	24	25	1	1	0.4096	1.64
	24	25	1	1	0.4096	1.64
	25	25	2	1	2.6896	1.64
	24	24	1	0	0.4096	0.08
	25	25	2	1	2.6896	1.64
	23	23	0	-1	0.1296	0.52
	24	24	1	0	0.4096	0.08
	23	24	0	0	0.1296	0.08
	23	25	0	1	0.1296	1.64
	24	24	1	0	0.4096	0.08
	23	25	0	1	0.1296	1.64
	24	23	1	-1	0.4096	0.52
	24	23	1	-1	0.4096	0.52
	23	24	0	0	0.1296	0.08
	24	23	1	-1	0.4096	0.52
Mean	23.4	23.72	0 total		63.52	110.08
SD	1.14	1.50				
n1	50					
n2	50					

Standard Deviation

S=sq.rt of((sum of d1square)+(sum of d2square))/n1+n2-2

(sum of d1square)+(sum of d2square) = 173.60

n1+n2-2 = 98

1.771429

S= 1.33095

t = (Mean X1 - Mean X2/S)(sqrt of n1n2/n1+n2)

Mean X1 - Mean X2/S = -0.270483 0.86

sqrt of n1n2/n1+n2 = 25

5

t= -1.352417 t value be consider as positive

Session : May 2009

APPENDIX – C

Table of Critical Values for T

	Two Tailed Significance							
	0.2	0.1	0.05	0.01	0.005	0.001	0.0005	0.0001
2	1.89	2.92	4.30	9.92	14.09	31.60	44.70	100.14
3	1.64	2.35	3.18	5.84	7.45	12.92	16.33	28.01
4	1.53	2.13	2.78	4.60	5.60	8.61	10.31	15.53
5	1.48	2.02	2.57	4.03	4.77	6.87	7.98	11.18
6	1.44	1.94	2.45	3.71	4.32	5.96	6.79	9.08
7	1.41	1.89	2.36	3.50	4.03	5.41	6.08	7.89
8	1.40	1.86	2.31	3.36	3.83	5.04	5.62	7.12
9	1.38	1.83	2.26	3.25	3.69	4.78	5.29	6.59
10	1.37	1.81	2.23	3.17	3.58	4.59	5.05	6.21
11	1.36	1.80	2.20	3.11	3.50	4.44	4.86	5.92
12	1.36	1.78	2.18	3.05	3.43	4.32	4.72	5.70
13	1.35	1.77	2.16	3.01	3.37	4.22	4.60	5.51
14	1.35	1.76	2.14	2.98	3.33	4.14	4.50	5.36
15	1.34	1.75	2.13	2.95	3.29	4.07	4.42	5.24
16	1.34	1.75	2.12	2.92	3.25	4.01	4.35	5.13
17	1.33	1.74	2.11	2.90	3.22	3.97	4.29	5.04
18	1.33	1.73	2.10	2.88	3.20	3.92	4.23	4.97
19	1.33	1.73	2.09	2.86	3.17	3.88	4.19	4.90
20	1.33	1.72	2.09	2.85	3.15	3.85	4.15	4.84
21	1.32	1.72	2.08	2.83	3.14	3.82	4.11	4.78
22	1.32	1.72	2.07	2.82	3.12	3.79	4.08	4.74
23	1.32	1.71	2.07	2.81	3.10	3.77	4.05	4.69
24	1.32	1.71	2.06	2.80	3.09	3.75	4.02	4.65
25	1.32	1.71	2.06	2.79	3.08	3.73	4.00	4.62
26	1.31	1.71	2.06	2.78	3.07	3.71	3.97	4.59
27	1.31	1.70	2.05	2.77	3.06	3.69	3.95	4.56
28	1.31	1.70	2.05	2.76	3.05	3.67	3.93	4.53
29	1.31	1.70	2.05	2.76	3.04	3.66	3.92	4.51

30	1.31	1.70	2.04	2.75	3.03	3.65	3.90	4.48
35	1.31	1.69	2.03	2.72	3.00	3.59	3.84	4.39
40	1.30	1.68	2.02	2.70	2.97	3.55	3.79	4.32
45	1.30	1.68	2.01	2.69	2.95	3.52	3.75	4.27
50	1.30	1.68	2.01	2.68	2.94	3.50	3.72	4.23
55	1.30	1.67	2.00	2.67	2.92	3.48	3.70	4.20
60	1.30	1.67	2.00	2.66	2.91	3.46	3.68	4.17
65	1.29	1.67	2.00	2.65	2.91	3.45	3.66	4.15
70	1.29	1.67	1.99	2.65	2.90	3.43	3.65	4.13
75	1.29	1.67	1.99	2.64	2.89	3.42	3.64	4.11
80	1.29	1.66	1.99	2.64	2.89	3.42	3.63	4.10
85	1.29	1.66	1.99	2.63	2.88	3.41	3.62	4.08
90	1.29	1.66	1.99	2.63	2.88	3.40	3.61	4.07
95	1.29	1.66	1.99	2.63	2.87	3.40	3.60	4.06
100	1.29	1.66	1.98	2.63	2.87	3.39	3.60	4.05
200	1.29	1.65	1.97	2.60	2.84	3.34	3.54	3.97
500	1.28	1.65	1.96	2.59	2.82	3.31	3.50	3.92
1000	1.28	1.65	1.96	2.58	2.81	3.30	3.49	3.91
Infinity	1.28	1.64	1.96	2.58	2.81	3.29	3.48	3.89

SESSION : MAY 2009

TABLE 39. TABLE OF THE DISTRIBUTION OF t FOR CERTAIN PROBABILITY LEVELS*

n	$P=.9$	.8	.7	.6	.5	.4	.3	.2	.1	.05	.02	.01	.001
1	.158	.325	.510	.727	1.000	1.376	1.963	3.078	6.314	12.706	31.821	63.657	636.619
2	.142	.289	.445	.617	.816	1.061	1.386	1.886	2.920	4.303	6.965	9.925	31.598
3	.137	.277	.424	.584	.765	.978	1.250	1.638	2.353	3.182	4.541	5.841	12.941
4	.134	.271	.414	.569	.741	.941	1.190	1.533	2.132	2.776	3.747	4.604	8.610
5	.132	.267	.408	.559	.727	.920	1.156	1.476	2.015	2.571	3.365	4.032	6.859
6	.131	.265	.404	.553	.718	.906	1.134	1.440	1.943	2.447	3.143	3.707	5.959
7	.130	.263	.402	.549	.711	.896	1.119	1.415	1.895	2.365	2.998	3.499	5.405
8	.130	.262	.399	.546	.706	.889	1.108	1.397	1.860	2.306	2.896	3.355	5.041
9	.129	.261	.398	.543	.703	.883	1.100	1.383	1.833	2.262	2.821	3.250	4.781
10	.129	.260	.397	.542	.700	.879	1.093	1.372	1.812	2.228	2.764	3.169	4.587
11	.129	.260	.396	.540	.697	.876	1.088	1.363	1.796	2.201	2.718	3.106	4.437
12	.128	.259	.395	.539	.695	.873	1.083	1.356	1.782	2.179	2.681	3.055	4.318
13	.128	.259	.394	.538	.694	.870	1.079	1.350	1.771	2.160	2.650	3.012	4.221
14	.128	.258	.393	.537	.692	.868	1.076	1.345	1.761	2.145	2.624	2.977	4.140
15	.128	.258	.393	.536	.691	.866	1.074	1.341	1.753	2.131	2.602	2.947	4.073
16	.128	.258	.392	.535	.690	.865	1.071	1.337	1.746	2.120	2.583	2.921	4.015
17	.128	.257	.392	.534	.689	.863	1.069	1.333	1.740	2.110	2.567	2.898	3.965
18	.127	.257	.392	.534	.688	.862	1.067	1.330	1.734	2.101	2.552	2.878	3.922
19	.127	.257	.391	.533	.688	.861	1.066	1.328	1.729	2.093	2.539	2.861	3.883
20	.127	.257	.391	.533	.687	.860	1.064	1.325	1.725	2.086	2.528	2.845	3.850
21	.127	.257	.391	.532	.686	.859	1.063	1.323	1.721	2.080	2.518	2.831	3.819
22	.127	.256	.390	.532	.686	.858	1.061	1.321	1.717	2.074	2.508	2.819	3.792
23	.127	.256	.390	.532	.685	.858	1.060	1.319	1.714	2.069	2.500	2.807	3.767
24	.127	.256	.390	.531	.685	.857	1.059	1.318	1.711	2.064	2.492	2.797	3.745
25	.127	.256	.390	.531	.684	.856	1.058	1.316	1.708	2.060	2.485	2.787	3.725
26	.127	.256	.390	.531	.684	.856	1.058	1.315	1.706	2.056	2.479	2.779	3.707
27	.127	.256	.389	.531	.684	.855	1.057	1.314	1.703	2.052	2.473	2.771	3.690
28	.127	.256	.389	.530	.683	.855	1.056	1.313	1.701	2.048	2.467	2.763	3.674
29	.127	.256	.389	.530	.683	.854	1.055	1.311	1.699	2.045	2.462	2.756	3.659
30	.127	.256	.389	.530	.683	.854	1.055	1.310	1.697	2.042	2.457	2.750	3.646
40	.126	.255	.388	.529	.681	.851	1.050	1.303	1.684	2.021	2.423	2.704	3.551
60	.126	.254	.387	.527	.679	.848	1.046	1.296	1.671	2.000	2.390	2.660	3.460
120	.126	.254	.386	.526	.677	.845	1.041	1.289	1.658	1.980	2.358	2.617	3.373
∞	.126	.253	.385	.524	.674	.842	1.036	1.282	1.645	1.960	2.326	2.576	3.291

* From Fisher and Yates, *Statistical Tables for Biological, Medical and Agricultural Research*, Edinburgh, Oliver and Boyd, Ltd., 1938, by permission of the publishers.

Assessment form (for examiner use only)

Candidate session number	0	0							

		Achievement level		
		First examiner	maximum	Second examiner
Assessment criteria	**A** research question	2	2	2
	B introduction	2	2	2
	C investigation	4	4	3
	D knowledge and understanding	4	4	4
	E reasoned argument	4	4	4
	F analysis and evaluation	3	4	4
	G use of subject language	4	4	4
	H conclusion	1	2	1
	I formal presentation	4	4	4
	J abstract	2	2	2
	K holistic judgment	3	4	4
	Total out of 36	33		34

Name of first examiner: _____ Examiner number: _____
(CAPITAL letters)

Name of second examiner: _____ Examiner number: _____
(CAPITAL letters)