BIBLIOGRAPHY

AGUSTIN, FLORESCA T. MAY 2007. Evaluating the Biofumigation Potentials of Various Brassica Species for the Control of *Ralstonia solanacearum* (E.F. Smith) Yabuuchi *et al.* Affecting Potatoes. Benguet State University, La Trinidad, Benguet.

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ABSTRACT

The application of brassica reduced the population of *Ralstonia solanacearum* in the soil with broccoli as the most effective biofumigant. However, the use of cabbage cv. scorpio significantly reduced the bacterial population. The use of chopped leaves and whole leaves effected the greatest reduction in the bacterial population. Among plant tissues, leaves effected a higher reduction in *R. solanacearum* population. In reducing bacterial population, the best combinations of biofumigant and tissue preparation were chopped leaves of broccoli, whole leaves of cauliflower, macerated roots and stems of cabbage cv. scorpio, and macerated leaves of mustard. The bacterial population in the untreated soil increased continuously over the sampling period.

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INTRODUCTION

Potato (*Solanum tuberosum* Linn.) ranks fourth among the most valuable crops grown worldwide. It provides of low-cost energy to the human diet and is a good source of carbohydrates, starch, proteins minerals, vitamins A and B_2 and some elements such as potassium and phosphorus (Kipps, 1979). In the Philippines, potato ranks third in production among the leading commercial vegetable crops and first among the vegetables in Northern Luzon. At present, 75% of the total production in the Cordillera region is from the province of Benguet. The suitability of potato production in Benguet (1600-2300 meters ASL) gives its estimated yield at 15.10 MT/ha (HARRDEC, 1996 as cited by Lando, 2002). However, this is seldom met due to losses from the occurrence of pest and diseases.

Bacterial wilt (BW) caused by *Ralstonia solanacearum* (E.F. Smith) Yabucchi *et al*. is the most serious soil-borne disease of solanaceous crops such as potato, tobacco, tomato and eggplant causing losses in quality and quantity of production (ACIAR, 2000). Perez *et al*. (1997) have shown that the disease has become severe in the highland and lowland potato production areas. *R. solanacearum* is capable of enduring in the soil and has a wide host range (Urquhart and Mienie, 1997 as cited by Lando, 2002). The host range includes 55 plant families especially members of *Solanaceae*.

Control measures are usually done through crop rotation, strict sanitation, use of resistant cultivars, use of disease-free planting material, and minimum tillage. Biofumigation using brassica tissues also decreases the incidence and severity of bacterial wilt (ACIAR, 2000). However, using different combination of control measures is more preferred to lessen the impact of the pathogen.

Biofumigation refers to the suppression of soil-borne pests and pathogens by naturally-occurring biocidal compounds, principally isothiocyanates (ITCs), released into the soil from decomposing organic material (ACIAR, 2000). Isothiocyanate compounds are similar to the methyl isothiocyanate (MITC) toxin from the metham sodium soil fumigant. These are highly biocidal to a diverse range of organisms including bacteria, nematodes, fungi, insects, and germinating seeds (Brown and Morra, 1997; Kirkegaard *et al.*, 1994; Potter *et al*,, 1998 and Walker, 1997). Thus, biofumigation may provide an option for farmers to manage BW based on their circumstances of disease pressure, economics and their ideals for cropping system.

Bacterial wilt is difficult to manage because it is soil-borne and seed-borne and there is no readily available chemical control. There is also great variation in the bacterium often manifested in a wide host range. Severely infested soils are not planted to potato anymore or are used in other industrial purposes. Thus, further reducing the potential potato production.

Biofumigation potentially provide a sustainable disease control option, for integrated BW management system while simultaneously improving soil health (soil fertility). The incorporation of biofumigants into the soil provides valuable organic matter, possibly reducing the dependence on organic fertilizers. Other benefits of biofumigation include improved soil texture, increased water holding capacity and improved microbial community structure (Harvey and Sams, 1999). The results of this study would help seed companies to develop biofumigant-type crucifers. Finally would open up opportunities in controlling other soil-borne pest and pathogens of other crop.

This study therefore aimed to evaluate the biofumigation potential of brassica for the control of *R. solanacearum* population in the soil, identify the brassica which is most effective biofumigant and determine the best method of brassica tissue preparation for biofumigation.

The study was conducted at the Department of Plant Pathology Laboratory and Greenhouse, Benguet State University, La Trinidad, Benguet from June to December 2005.

REVIEW OF LITERATURE

The Disease

Bacterial wilt (BW) is one of the most important, widespread and lethal bacterial diseases of plants (Ma, 1990 as cited by Lando, 2002). It is considered a dreaded disease in many parts of Asia, Africa, and Central and South America. In 2000, the Australia Centre for International Agricultural Research (ACIAR) recognized it as one of the most important diseases of bacterial origin in the world. Bacterial wilt is the number two constraint on potato production in over 40 developing countries (CGIAR, 2005).

Hayward in 1985 reported that more than 55 crops and wild species are affected by R. *solanacearum* crops such as potato, tobacco, tomato, eggplant, banana, chili, bell pepper and peanut are highly susceptible to the disease. Recently it was shown that certain ecotypes of the model plant *Arabidopsis thaliana* are also susceptible to the pathogen (Hayward, 2000). An endemic strain (race 1, biovar 1) was detected in 2001 in geraniums (*Pelargonium* sp.) in Florida, USA (Momol *et al*., 2003). It is common in tropical, subtropical and warm temperature regions where temperature and moisture conditions are favorable for its development (Singh, 1978). The bacterium may also be present in cooler climates such as relatively high elevation in the tropics or higher latitudes.

The Causal Organism

The bacterium responsible for bacterial wilt is *Ralstonia solanacearum* (E. F. Smith) Yabucchi *et al*., (1996) formerly known as *Pseudomonas solanacearum* E. F. Smith. It is a Gram negative, strictly aerobic, nospore-forming, noncapsulate, nitrate

reducing, catalase-positive, ammonia-forming, and monotrichous short rod (1.5 x 0.5 mm) (Sands *et al*., 1980; Stanier *et al*., 1966). The wild type bacterium is usually nonmotile and does not form a polar flagellum in a liquid medium. Avirulent variants that develop in culture are actively motile. Colonies on agar are opalescent which became darker with age. Rich (1983) described them as small, irregular, smooth, wet and shiny. The virulent colonies are pink in tetrazolium chloride agar (TCZA). The optimum temperature for growth ranges from 35^0C- 37^0C (Weber, 1973 and Rich, 1983) and the thermal death point lies at about 52^0C (Kerr, 1983).

R. *solanacearum* cannot hydrolyze starch and it can liquefy gelatin slowly or not at all. The bacterium is inhibited by relatively low concentrations of salt in broth culture and is sensitive to desiccation.

The cultured bacterium in unaerated liquid media losses its virulence and viability rapidly and change from the fluidal (nonmotile) wild type to the avirulent, highly motile variants.

Symptoms

Bacterial wilt is described by Agrios (1997) as sudden wilt. Infected plants die rapidly. Older leaves may first show leaf dropping and discoloration or one-sided wilting and stunting before completely wilts and permanently dies. Severely infected tubers are blackened and when cut, vascular ring turns brown. In general, the symptoms are wilting, stunting and yellowing of the leaves, followed by the collapse of the entire plants (Agrios, 1978).

Symptoms occur both above and belowground parts of the host plants. Above ground symptoms include wilting, stunting, and yellowing of leaves. Infection is

characterized by initial wilting only of a part of the stems of the plant, or even one side of the leaf or stem. The entire plant wilts quickly without yellowing when development of the disease is rapid (French, 1996).

External symptoms on the tuber are visible at harvest when infection is severe. Bacterial ooze collects at tuber eyes causing soil to adhere (CIP, 2004). Brownish discolorations on the vascular rings are observed. When tubers are slightly squeezed, ooze comes out naturally from the rings. In more advance stages of the disease development, the vascular ring or the whole tuber may disintegrate completely (Bahar and Danish, 1990; Martin and French, 1996).

Survival

Bacteria live in follow soil for 6 years or more and may persist indefinitely in the presence of susceptible plants (Pope, 1995). R. *solanacearum* is capable of surviving under high moisture. This proves the field observation that BW is more serious in wet, humid, tropical areas than in desert areas, even under irrigation (Buddenhagen and Kelman, 1964). However, survival depends on race involved and depends on deep soil layer (Persley, 1995).

Management

French (1996) stated that the inherent variability of R. *solanacearum* and the strong influence of environmental conditions on resistance make disease management difficult. In managing the disease, crop rotation, prevention and use of resistant cultivars are employed (Geesteranus as cited by Lando, 2002).

Using areas free of the bacterium reduces the chance of tubers of becoming infected later. In a field infested with BW it takes at least 2 years rotation with non-susceptible crops to decrease its population. Planting pathogen-free seed tubers will decrease severity and incidence in infested field and will prevent the introduction of the pathogen into non-infected area. The elimination of alternate hosts can decrease pathogen population inoculum in the soil. The use of resistant cultivars play an important role but the extreme variability found in *R. solanacearum* makes breeding for resistance to the pathogen difficult (Sequeria, 1983).

Biofumigation

Biofumigation has been shown to reduce the levels of several soil pathogens, including bacterial wilt and root knot nematode. Isothiocyanates (ITCs) were released from brassica tissues when glucosinolates (GSLs) are hydrolyzed by endogenous myrosinase enzyme (Angus *et al*., 1994). These hydrolysis products (ITCs) are known to have broad biocidal activity including insecticidal, nematicidal, fungicidal, antibiotic and phytotoxic effects (reviewed by Brown and Morra, 1997). The difference in structure of individual GSLs and ITCs depends on their organic side-chain (aliphatic, aromatic or indole). Their concentrations, profiles, distribution and toxicity varies within and between brassica species and in different plant tissues, hence, the concentration and type of biocidal hydrolysis product involved also varies (Kirkegaard and Sarwar, 1998).

Biofumigation is one of the newest technology as a component of integrated pest management. Potential of brassicas (ITCs) were observed during the early 1990s when wheat crops grew vigorously following brassica break crops such as canola and Indian mustard than other break crops such as linseed or oats (Angus *et al*., 1991; Kirkegaard *et*

al., 1994). In the study of Kirkegaard *et al*. (1998), they grew wheat in pots inoculated with the take-all fungus (*Gaeumannomyces graminis* var. *tritici*) and incorporated the root residues of canola. Results showed lower take- all infection on wheat after brassicas compared to linseed. Aromatic GSL present in canola roots have been shown to be highly toxic to cereal fungal pathogens and genetic diversity within Australian canola varieties allows selection of higher root GSL levels (Kirkegaard and Sarwar, 1999).

The work of Harding and Wicks (1999) shows that ITCs emanating from *Brassica juncea* (Indian mustard) meal, B. *juncea*/B. *napus* (canola) leaves and roots are inhibitory to a number of significant fungal pathogens of potato (*Rhizoctonia* $AG_{3,}$ *Rhizoctonia* $AG_{8,}$ *Verticillium dahliae* (A and B), *Colletotrichum coccodes* (A and B), *Phytophthora erythroptica* and *P*. *coryptogea*).

The preliminary studies on bacterial wilt caused by R. *solanacearum* indicate that tissues of mustards (B. *juncea*, B. *nigra* and B. *carimata*) are more suppressive than those of other brassicas tested suggesting that 2-propenyl GSL is the active compound (Akiew, unpublished). This compound is also present in kale and cabbage indicating residues from these crops may also have activity against BW. The results of the study of Akiew in 1999 demonstrate reductions of 40-80% in BW in the field by Indian mustard green manure and up to 97% reduction in the glasshouse when combined with organic soil amendments. A commercial Indian biofumigant (FUMUS) showed potential to reduce levels of BW and increase potato yield dramatically (from 0.3-22 tons/ha) in an on-farm trial in Victoria (ACIAR, 2000; Akiew *et al*., 1996).

In an experiment at Southedge Research Station BW nursery, Australia, the radish, fodder rape and mustard biofumigants were all effective in delaying disease onset,

reducing the incidence and severity of BW and increasing eggplant yield (ACIAR, 2000). Incorporation of some brassica fumigants has reduced BW by 50-60-% in three of the four experiments of ACIAR in 2002 compared to non – brassica controls.

In north Queensland, Australia, results in some of the field trials have been excellent. A paddock with high-level infection of bacterial wilt was planted with tomatoes. An untreated block yielded less than two tons of tomatoes, while the area where a brassica green manure treatment had been applied yielded up to 20 tons of tomatoes and had correspondingly lower levels of bacterial wilt (Taylor, 2006).

In the Philippines, trials have been planted at a range of field sites, from high-elevation areas in Benguet, with cabbages, potatoes and other temperate crops, down to lowland areas in Mindanao, where eggplants and tomatoes are the major crops. According to Kirkegaard (2006) the most promising treatments (radish, mustard and broccoli) have reduced bacterial wilt significantly (50 to 60%) in most of the experiments.

MATERIALS AND METHODS

Sources of Test Materials

Ralstonia solanacearum was isolated from naturally infected potatoes and pure culture was prepared following standard procedures. Brassicas were collected from the fields in Atok, Kabayan, La Trinidad, and Tuba, Benguet.

Preparation of Brassica Tissues

Brassica plants were collected. Soil was washed from the roots of all plants and subsamples from each sample were separated into stem and root tissue and leaf tissue. Macerated specimens were first cut into pieces then blended, chopped specimens were cut into at least 2-3 cm long pieces and whole tissue specimens were used as is.

Each treatment was replicated thrice and arranged in a factorial completely randomized design (CRD) with plant species as factor A and the different type of plant tissue and method of preparation as factor B. The treatments were as follows:

Factor A	Plant Species
A_0	Untreated (Control 1)
A_1	Sunflower - Control 2 (*Tithonia diversifolia*)
A_2	Broccoli cv. Marathon (*Brassica oleracea var. italica*)
A_3	Cauliflower cv. Milkyway (*Brassica oleracea var. botrytis*)
A_4	Cabbage cv. Scorpio (*Brassica oleracea var. capitata*)
A_5	Cabbage cv. Rareball (*Brassica oleracea var. capitata*)
A_6	Radish cv. Mino early (*Raphanus sativa*)
A_7	Mustard cv. Montana (*Brassica juncea*)

A_8 Pechay (*Brassica pekenensis*)

A_9 Radish + Mustard (1:1)

Factor B Types of plant tissue and methods of preparation

B_1 Macerated stems and roots

B_2 Macerated leaves

B_3 Chopped roots and stems

B_4 Chopped leaves

B_5 Whole roots and stems

B_6 Whole leaves

Evaluation of the Bactericidal Potential of Brassica Tissues

Laboratory Experiment. One hundred ml of bacterial suspension was incorporated with 200 g sterile soil in plastic cups and allowed to stand for two days. A 10 - g soil sample was then taken to determine initial population (cfu/g soil) of the bacterium and Brassica tissues (10g/200g of soil) were incorporated. Bacterial population in the soil was monitored weekly for five weeks following standard procedures.

Data Gathered

The following were the data gathered/computed:

1. Colony counts. Colonies that shows the typical characteristics of the bacterium were counted.
2. cfu/g soil was calculated as follows:

$$\text{cfu/g soil} = \frac{\frac{\text{average colony count x DF}}{\text{amount plated}}}{\text{10 g soil}}$$

a. Initial population (cfu/g soil). The initial population of the bacterium in the soil was measured two days after inoculation.

b. Weekly population (cfu/g soil). Soil samples were obtained from the treatments every week for five weeks.

c. Final population (cfu/g soil). Final population was taken on the fifth week from inoculation.

RESULTS AND DISCUSSIONS

Effect of Biofumigant

After seven days from incorporation, the brassica biofumigants had an immediate effect in reducing the *Ralstonia solanacearum* population except for sunflower, cabbage and pechay in which there was an increase in bacterial population (Table 1). Generally, the bacterial population decreases with the application of biofumigants, however there were slight increase observed during the second to the fourth assessment period. Results revealed that the application of broccoli consistently decrease R. *solanacearum* population in the soil. On the other hand, the bacterial population in the soil treated with pechay consistently increase. On the final week, there was a decrease in R. *solanacearum* population in most of the treatments. Greatest reduction in bacterial population was observed in the soil treated with broccoli followed by cabbage cv. scorpio, radish + mustard, cabbage cv. rareball and cauliflower (Figure 1). There was an increase in bacterial population in the untreated soil and the soil treated with pechay and radish. This shows that pechay had no impact in reducing R. *solanacearum* population. Pechay decomposed much faster than the other biofumigants. The ease by which pechay decomposed may have caused the release of ITC's much faster than the other treatments, thereby causing the faster dissipation of any biofumigant content. It was also the findings of ACIAR that pechay had no effect on the reduction of R. *solanacearum* population. The application of radish show a negative effect in the reduction of bacterial population, this proves that GSL content varies within brassica varieties (ACIAR, 2000).

Table 1. Effect of biofumigant on the population of R. *solanacearum* (log cfu x 10^4)

BIOFUMIGANT	ASSESSMENT PERIOD					
	Initial	Week 1	Week 2	Week 3	Week 4	Week 5
Sunflower	0.6^{bc}	0.76^{a}	0.84^{ab}	0.61^{ab}	0.54^{c}	0.52^{bc}
Broccoli	0.61^{bc}	0.52^{b}	0.43^{d}	0.41^{c}	0.32^{d}	0.29^{e}
Cauliflower	0.68^{b}	0.57^{b}	0.67^{bc}	0.55^{ab}	0.38^{d}	0.44^{cd}
Cabbage cv. Scorpio	0.59^{bc}	0.66^{ab}	0.78^{ab}	0.62^{ab}	0.56^{bc}	0.32^{de}
Cabbage cv. Rareball	0.42^{d}	0.57^{b}	0.94^{a}	0.67^{a}	0.68^{ab}	0.43^{cd}
Radish	0.56^{c}	0.52^{b}	0.69^{bc}	0.56^{ab}	0.77^{a}	0.62^{ab}
Mustard	0.94^{a}	0.79^{a}	0.68^{bc}	0.62^{ab}	0.58^{bc}	0.65^{a}
Pechay	0.53^{c}	0.56^{b}	0.59^{cd}	0.66^{ab}	0.67^{ab}	0.68^{a}
Radish + Mustard	0.68^{b}	0.6^{b}	0.55^{cd}	0.54^{b}	0.43^{d}	0.41^{cd}
Untreated	0.95^{a}	0.92^{a}	0.93^{a}	1.06^{a}	1.23^{a}	1.31^{a}

The ITCs produced vary between brassica species and toxicity may sometimes differ among organisms (Brown and Morra, 1997). The level of glucosinolates in broccoli is higher than in other crucifers and was highly biocidal to a diverse range of organisms including nematodes, bacteria, fungi, insects and germinating seeds (Kirkegaard *et al.*, 1994 and Brown and Morra, 1997). The positive effect of sunflower show, on the other hand, that even nonbrassicaceous organic matter may reduce R. *solanacearum* population. This may point to other non GSL-related effects caused by the decomposing organic matter.

Guilabo (2005) stated that broccoli and cauliflower decreased the population of R. *solanacearum* in the soil. In addition the application of cauliflower effectively reduced

bacterial wilt incidence. Furthermore, Akiew (1999) cited that the application of mustard reduced bacterial wilt at 40-80 %. According to the ACIAR project on biofumigation the most promising brassica tested in the Philippines (using radish, mustard and broccoli) have reduced bacterial wilt by 50 to 60 %. The reduction in bacterial population reduces the inoculum in the soil and will in turn reduce the incidence and severity of disease.

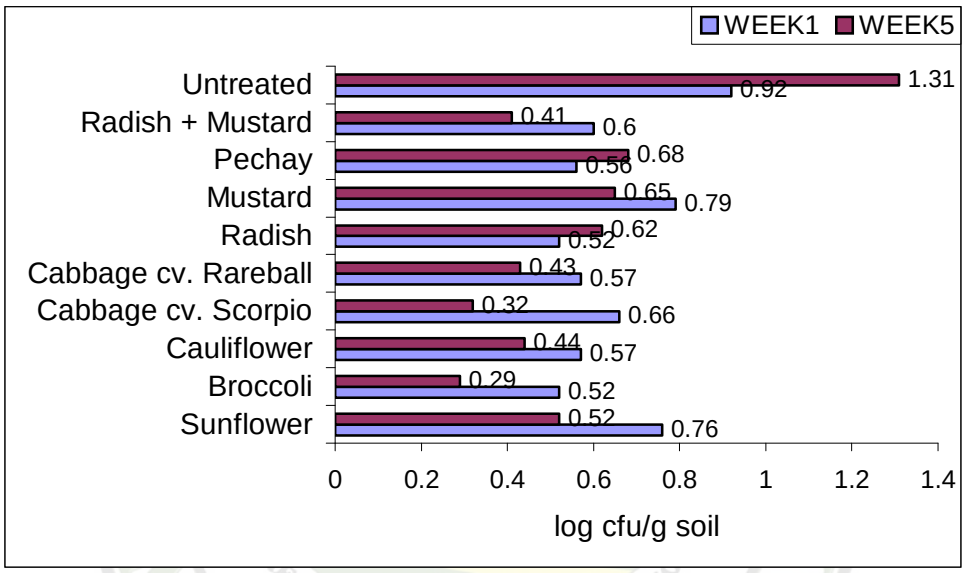

Figure 1. The change in the R. *solanacearum* population in the soil as affected by the biofumigants (week 1 and week 5)

Effect of Tissue Preparation

Generally, there was an increase in the population of *Ralstonia solanacearum* after seven days of incorporating biofumigants. However, immediate reduction in bacterial population was observed with the use of chopped roots and stems and whole leaves. The general trend from week 2 to week 4 was decrease of bacterial population although in some treatments there was a slight increase in population. On the final week, all the treatments had reduced *R. solanacearum* population in the soil (Table 2). The greatest reduction was effected by the application of chopped leaves but was not

significantly different from the use of whole leaves. Between the root and stem and leaf tissues, the leaf tissue effected the higher reduction of bacterial population (Figure 2). The leaves decomposed faster than the roots and stems, thus production of ITCs is also faster.

Table 2. Effect of tissue type and preparation on the population of R. *solanacearum* (log cfu x 10^4)

TISSUE TYPE AND PREPARATION	ASSESSMENT PERIOD					
	Initial	Week 1	Week 2	Week 3	Week 4	Week 5
Macerated roots and stems	0.54^{c}	0.68^{ab}	0.73^{a}	0.63^{ab}	0.53^{a}	0.51^{ab}
Macerated leaves	0.68^{ab}	0.71^{a}	0.76^{a}	0.47^{c}	0.55^{a}	0.52^{ab}
Chopped roots and stems	0.75^{a}	0.64^{ab}	0.64^{ab}	0.66^{a}	0.55^{a}	0.55^{a}
Chopped leaves	0.58^{c}	$0.6a^{bc}$	0.63^{ab}	0.59^{ab}	0.59^{a}	0.38^{c}
Whole roots and stems	0.51^{c}	0.56^{bc}	0.57^{b}	0.53^{bc}	0.63^{a}	0.51^{ab}
Whole leaves	0.68^{ab}	0.51^{c}	0.79^{a}	0.59^{ab}	0.47^{b}	0.44^{bc}

In general, green fresh leaf residues of brassica incorporated in the soil released greater amounts of ITCs and may suppress pests better than the dried or mature residues as those found in roots and stems. As Matthiessen *et al.* (2001) noted, aromatic ITCs produced from GSLs often found in roots are very toxic but they are of low volatility and that contact with organisms may be reduced. Aliphatic ITCs found mostly on shoots are less toxic but of greater volatility that allow easier contact with organisms. As shoot is generally very much greater than root biomass, shoots contribute more of the total ITC

potential (Matthiessen and Kirkegaard, 1998). ACIAR Project No. SMCN/200/114 points out that tissue disruption increases the efficiency of release of ITCs achieved by rough chopping and blending/ macerating. By chopping the plant cells, the vacuoles in which the GSLs are stored were destroyed and able to meet the myrosinase enzyme in the cytoplasm. However, macerated plant tissues in this experiment show a lower reduction in the bacterial population compared to chopped and whole plant tissues. This was maybe due to faster loss of isothiocyanates. Longer-term incubation (30 days) of tissues that were not macerated can sometimes effective as macerated amendments against BW (ACIAR, 2000). ACIAR (2000) thus concluded, "The general longer-term impacts of organic amendments can also play a role in pest suppression presumably due to induced changes in the soil microbial community favoring pest antagonist".

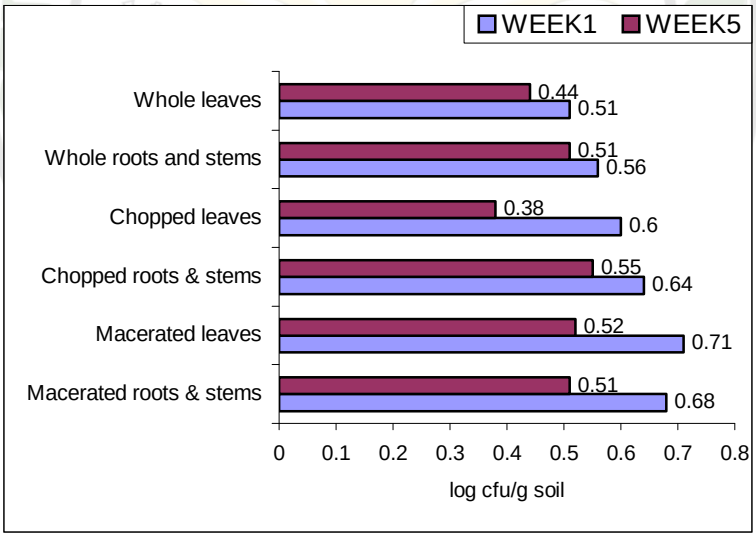

Figure 2**.** The change in the R. *solanacearum* population in the soil as affected by the tissue preparation (week 1 and week 5)

Interaction Effect

Figure 3 shows the decrease in the population of *R. solanacearum* in the selected best combinations of biofumigants and tissue preparation. The use of chopped leaves of broccoli, whole leaves of cauliflower, macerated roots of cabbage cv. scorpio, macerated leaves of mustard, whole leaves and chopped leaves of radish + mustard effected the greatest reduction. The summary of the changes in the population of *R. solanacearum* was presented in Figure 5.

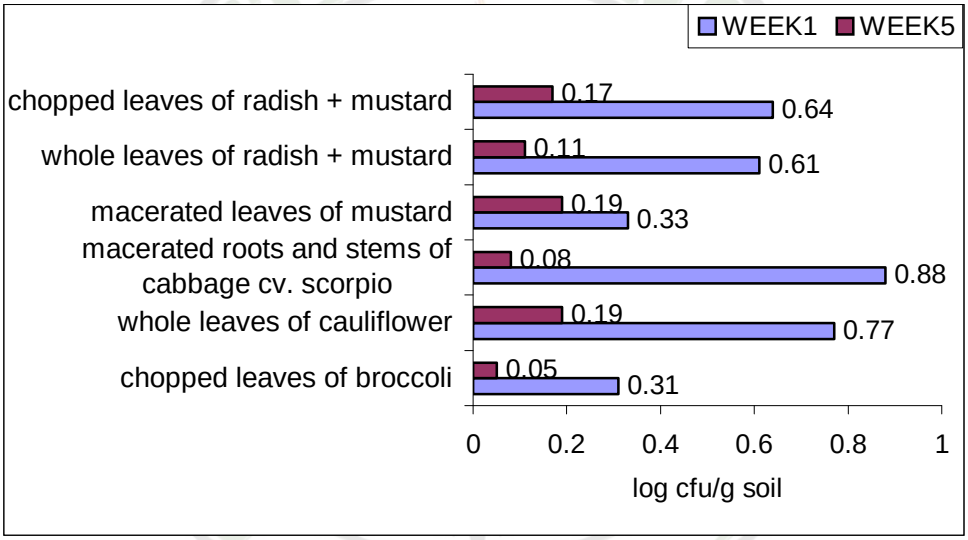

Figure 3. Decrease in the population of R. *solanacearum* in the selected best combinations of biofumigant and tissue preparation

The greatest increase in the bacterial population was observed in untreated soil, and in soil treated with whole roots and stems of cabbage cv. rareball, macerated roots and stems of sunflower, chopped roots and stems of sunflower, macerated leaves and whole leaves of pechay (Figure 4). The GSL types and concentrations vary within and between brassica species and in the different plant tissues (Brown and Morra, 1997). It is also affected by age, health and nutrition and edaphic environment in which tissues are

incorporated. The nutrients supplied to the plant during its growth affects the level of GSL. Efficiency of release of ITCs from incorporated tissues is also influence by the type of soil, moisture content and degree of tissue disruption.

These results further indicate the potential use of biofumigation for the control of *Ralstonia solanacearum*. The range in glucosinolate profiles, the differential toxicity of isothiocynate to different pest and the wide range in phenological and morphological diversity in brassica provides significant scope to select or breed brassicas with enhanced biofumigation potential for a particular target organism (Kirkegaard *et. al*., 1998).

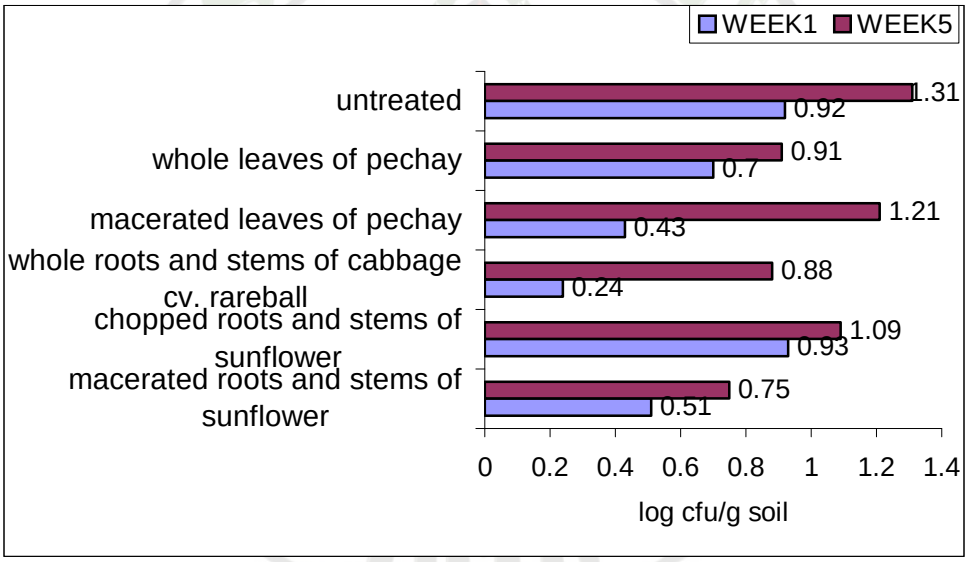

Figure 4. The greatest increase in the population of R. *solanacearum* as affected by the combinations of biofumigant and tissue preparation

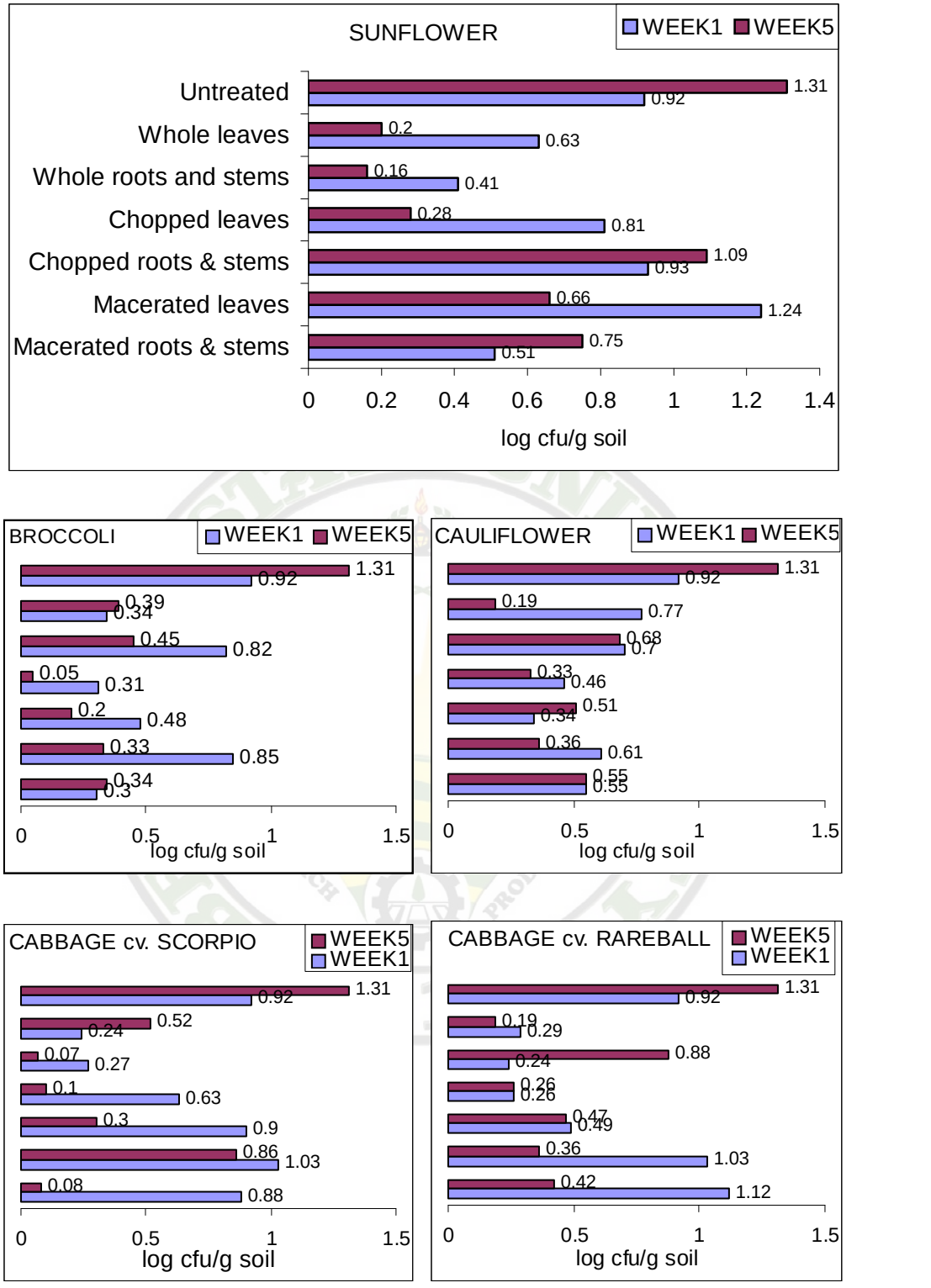

Figure 5a. Summary of the changes in the population of R. *solanacearum* as affected by the biofumigant and tissue preparation

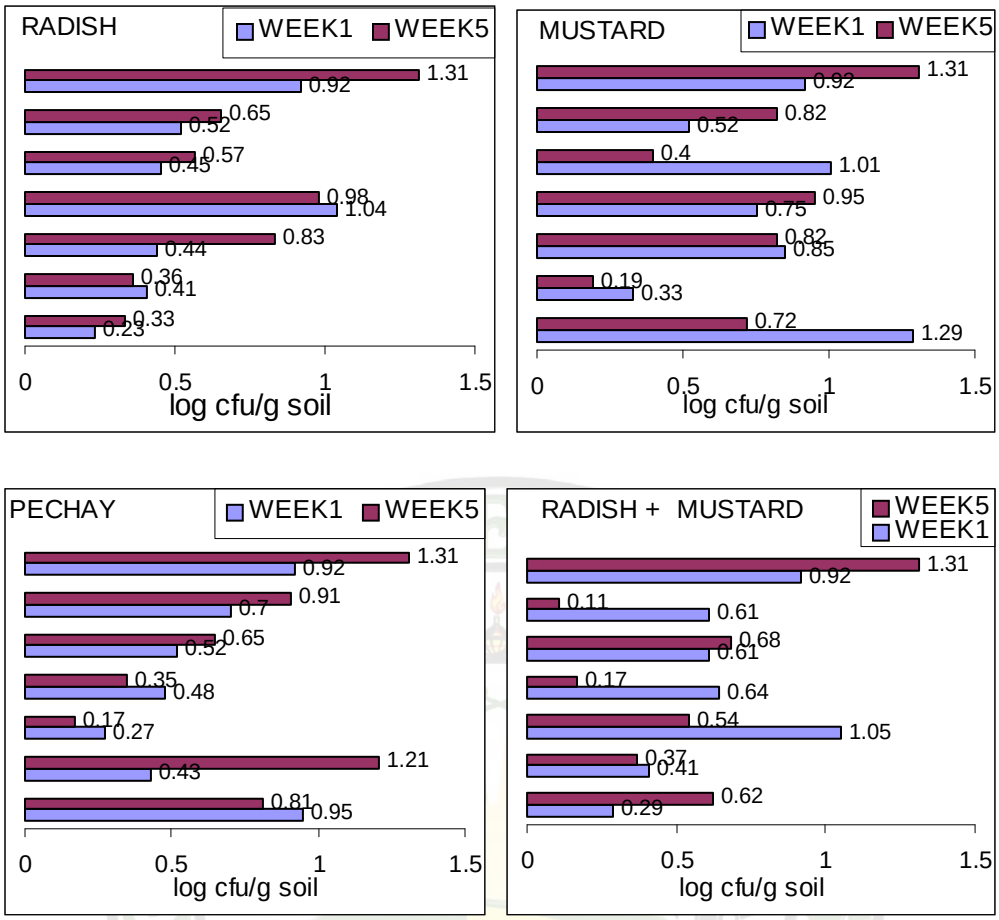

Figure. 5b. Summary of the changes in the population of R. *solanacearum* as affected by the biofumigant and tissue preparation

SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

Summary

The study was conducted at the laboratory and greenhouse of the Department of Plant Pathology, Benguet State University, La Trinidad, Benguet to evaluate the biofumigation potentials of various brassica species for the control of *Ralstonia solanacearum* (E. F. Smith) Yabuuchi *et al*. affecting potatoes.

Results showed that the application of brassica can effectively reduce the population of *Ralstonia solanacearum* in the soil with broccoli as the most effective biofumigant. However, the use of cabbage cv. scorpio significantly reduces the bacterial population. The use of chopped leaves and whole leaves effected the greatest reduction in the bacterial population. Among plant tissues, leaves effected a higher reduction in *R. solanacearum* population. In reducing bacterial population, the best combinations of biofumigant and tissue preparation were chopped leaves of broccoli, whole leaves of cauliflower, macerated roots and stems of cabbage cv. scorpio, and macerated leaves of mustard. The bacterial population in the untreated soil continuously increased throughout the sampling period.

Conclusions

Based on the findings of this study, biofumigants can be used to reduce the soil population of *Ralstonia solanacearum*. Furthermore, the incorporation of brassicaceous "waste" material is an economical and environmentally-safe practice that can be used by resource-poor farmers. Finally, the use of cruciferous biofumigants can be a viable

option in an integrated and long-term management of bacterial wilt disease both in conventional and in organic production systems.

Recommendations

The following are hereby recommended:

1. A follow-up study to confirm these results must be conducted.
2. A farm trial must be conducted to evaluate the efficacy of using biofumigants for the control of R. *solanacearum* in the field.
3. A study should be conducted to determine the effect of biofumigants to other soilborne pathogens.
4. Finally, inclusion of biofumigants in integrated bacterial wilt management should be considered.

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APPENDICES

Appendix Table 1**.** Initial population of *Ralstonia solanacearum* in the soil (log cfu/g soil x 10^4)

TREATMENT	REPLICATION I	II	III	TOTAL	MEAN
Sunflower					
Macerated root + stem	0.52	0.32	0.4	1.24	0.41
Macerated leaf	0.34	0.49	0.68	1.51	0.50
Chopped root + stem	0.81	0.86	0.91	2.58	0.86
Chopped leaf	0.67	0.58	0.74	1.99	0.66
Whole root + stem	0.56	0.43	0.32	1.31	0.44
Whole leaf	0.4	0.94	0.81	2.15	0.72
Broccoli					
Macerated root + stem	0.36	0.11	0.2	0.67	0.22
Macerated leaf	0.66	0.59	0.64	1.89	0.63
Chopped root + stem	0.92	0.85	1.14	2.91	0.97
Chopped leaf	0.77	0.2	0.23	1.2	0.40
Whole root + stem	0.7	0.41	0.67	1.78	0.59
Whole leaf	0.6	0.95	1.01	2.56	0.85
Cauliflower					
Macerated root + stem	0.4	0.68	0.45	1.53	0.51
Macerated leaf	1.15	1.06	0.61	2.82	0.94
Chopped root + stem	0.88	0.87	0.72	2.47	0.82
Chopped leaf	0.3	0.32	0.53	1.15	0.38
Whole root + stem	0.18	0.32	0.4	0.9	0.30
Whole leaf	1.03	1.1	1.09	3.22	1.07
Cabbage cv. Scorpio					
Macerated root + stem	0.67	0.36	0.4	1.43	0.48
Macerated leaf	0.8	0.49	1	2.29	0.76
Chopped root + stem	0.8	0.68	0.91	2.39	0.80
Chopped leaf	0.79	0.57	0.46	1.82	0.61
Whole root + stem	0.32	0.28	0.43	1.03	0.34
Whole leaf	0.64	0.45	0.51	1.6	0.53
Cabbage cv. Rareball					
Macerated root + stem	0.48	0.38	0.76	1.62	0.54
Macerated leaf	0.51	0.95	0.87	2.33	0.78
Chopped root + stem	0.38	0.48	0.52	1.38	0.46
Chopped leaf	0.08	0.38	0.26	0.72	0.24

Appendix Table 1. Continued...

TREATMENT	REPLICATION I	II	III	TOTAL	MEAN
Whole root + stem	0.36	0.2	0.28	0.84	0.28
Whole leaf	0.11	0.2	0.32	0.63	0.21
Radish					
Macerated root + stem	0.43	0.32	0.38	1.13	0.38
Macerated leaf	0.45	0.54	0.45	1.44	0.48
Chopped root + stem	0.41	0.58	0.62	1.61	0.54
Chopped leaf	0.93	1.13	1.08	3.14	1.05
Whole root + stem	0.32	0.57	0.52	1.41	0.47
Whole leaf	0.51	0.34	0.43	1.28	0.43
Mustard					
Macerated root + stem	1.45	1.06	1.23	3.74	1.25
Macerated leaf	0.91	0.79	0.9	2.6	0.87
Chopped root + stem	0.95	1.06	1.09	3.1	1.03
Chopped leaf	0.81	0.43	0.54	1.78	0.59
Whole root + stem	1.07	1	0.99	3.06	1.02
Whole leaf	0.91	0.97	0.68	2.56	0.85
Pechay					
Macerated root + stem	0.68	0.57	0.58	1.83	0.61
Macerated leaf	0.49	0.41	0.48	1.38	0.46
Chopped root + stem	0.4	0.26	0.36	1.02	0.34
Chopped leaf	0.45	0.48	0.49	1.42	0.47
Whole root + stem	0.69	0.56	0.34	1.59	0.53
Whole leaf	1	0.82	0.48	2.3	0.77
Radish + mustard					
Macerated root + stem	0.67	0.32	0.32	1.31	0.44
Macerated leaf	0.62	0.63	0.53	1.78	0.59
Chopped root + stem	1.08	0.84	0.99	2.91	0.97
Chopped leaf	0.95	0.51	0.88	2.34	0.78
Whole root + stem	0.73	0.49	0.68	1.9	0.63
Whole leaf	0.71	0.6	0.63	1.94	0.65
Untreated	0.9	1.01	0.94	2.85	0.95
TOTAL	34.81	31.78	33.94	100.53	
MEAN	0.65	0.59	0.63		0.62

ANOVA TABLE

SOURCE OF VARIATION	DF	SS	MS	FC	Tab F 0.05	Tab F 0.01
Factor A	8	2.90	0.36	16.92**	2.03	2.69
Factor B	5	1.21	0.24	11.33**	2.3	3.2
A x B	40	5.65	0.14	6.59**	1.51	1.79
Error	108	2.32	0.02			
TOTAL	161	12.08				

CV = 29.35%

Appendix Table 2**.** Week one population of *Ralstonia solanacearum* in the soil (log cfu/g soil x 10^4)

TREATMENT	REPLICATION I	II	III	TOTAL	MEAN
Sunflower					
Macerated root + stem	0.46	0.48	0.58	1.52	0.51
Macerated leaf	0.96	1.51	1.26	3.73	1.24
Chopped root + stem	0.76	1.01	1.03	2.8	0.93
Chopped leaf	0.6	0.81	1.03	2.44	0.81
Whole root + stem	0.3	0.61	0.32	1.23	0.41
Whole leaf	0.18	0.93	0.78	1.89	0.63
Broccoli					
Macerated root + stem	0.15	0.3	0.45	0.9	0.30
Macerated leaf	0.72	0.79	1.05	2.56	0.85
Chopped root + stem	0.36	0.4	0.67	1.43	0.48
Chopped leaf	0.3	0.26	0.36	0.92	0.31
Whole root + stem	0.45	1.03	0.98	2.46	0.82
Whole leaf	0.26	0.28	0.49	1.03	0.34
Cauliflower					
Macerated root + stem	0.46	0.58	0.6	1.64	0.55
Macerated leaf	0.59	0.72	0.51	1.82	0.61
Chopped root + stem	0.32	0.28	0.43	1.03	0.34
Chopped leaf	0.34	0.46	0.57	1.37	0.46
Whole root + stem	0.58	0.62	0.91	2.11	0.70
Whole leaf	0.79	0.92	0.59	2.3	0.77
Cabbage cv. Scorpio					
Macerated root + stem	1.32	0.34	0.97	2.63	0.88
Macerated leaf	1.43	0.88	0.79	3.1	1.03
Chopped root + stem	0.92	0.76	1.01	2.69	0.90
Chopped leaf	0.88	0.58	0.43	1.89	0.63
Whole root + stem	0.11	0.4	0.3	0.81	0.27
Whole leaf	0.08	0.26	0.38	0.72	0.24
Cabbage cv. Rareball					
Macerated root + stem	0.84	1.25	1.27	3.36	1.12
Macerated leaf	1.38	1.19	0.51	3.08	1.03
Chopped root + stem	0.32	0.51	0.65	1.48	0.49
Chopped leaf	0.28	0.28	0.23	0.79	0.26
Whole root + stem	0.11	0.2	0.4	0.71	0.24
Whole leaf	0.18	0.45	0.23	0.86	0.29

Appendix Table 2**.** Continued...

TREATMENT	REPLICATION I	II	III	TOTAL	MEAN
Radish					
Macerated root + stem	0.23	0.15	0.3	0.68	0.23
Macerated leaf	0.26	0.58	0.4	1.24	0.41
Chopped root + stem	0.45	0.54	0.34	1.33	0.44
Chopped leaf	0.99	1.05	1.09	3.13	1.04
Whole root + stem	0.49	0.41	0.46	1.36	0.45
Whole leaf	0.52	0.61	0.43	1.56	0.52
Mustard					
Macerated root + stem	1.29	1.32	1.27	3.88	1.29
Macerated leaf	0.3	0.41	0.28	0.99	0.33
Chopped root + stem	0.6	0.95	1.01	2.56	0.85
Chopped leaf	0.63	1.03	0.59	2.25	0.75
Whole root + stem	1.37	0.78	0.88	3.03	1.01
Whole leaf	0.43	0.45	0.68	1.56	0.52
Pechay					
Macerated root + stem	0.9	1.03	0.92	2.85	0.95
Macerated leaf	0.3	0.59	0.41	1.3	0.43
Chopped root + stem	0.36	0.26	0.18	0.8	0.27
Chopped leaf	0.45	0.46	0.52	1.43	0.48
Whole root + stem	0.7	0.32	0.53	1.55	0.52
Whole leaf	0.86	0.72	0.52	2.1	0.70
Radish + mustard					
Macerated root + stem	0.15	0.32	0.41	0.88	0.29
Macerated leaf	0.38	0.49	0.36	1.23	0.41
Chopped root + stem	0.86	1	1.28	3.14	1.05
Chopped leaf	0.08	0.34	1.49	1.91	0.64
Whole root + stem	0.59	0.64	0.59	1.82	0.61
Whole leaf	0.69	0.53	0.62	1.84	0.61
Untreated	0.88	1.24	0.63	2.75	0.92
TOTAL	27.56	30.75	30.59	88.9	
MEAN	0.51	0.57	0.57		0.55

ANOVA TABLE

SOURCE OF VARIATION	DF	SS	MS	FC	Tab F 0.05	Tab F 0.01
Factor A	8	1.44	0.18	3.92**	2.03	2.69
Factor B	5	0.72	0.14	3.14*	2.3	3.2
A x B	40	10.92	0.27	5.94**	1.51	1.79
Error	108	4.96	0.05			
TOTAL	161	18.05				

CV = 34.82%

Appendix Table 3. Week two population of *Ralstonia solanacearum* in the soil (log cfu/g soil x 10^4)

TREATMENT	REPLICATION I	II	III	TOTAL	MEAN
Sunflower					
Macerated root + stem	1.27	1.18	1.1	3.55	1.18
Macerated leaf	0.71	0.76	0.67	2.14	0.71
Chopped root + stem	0.82	1.08	0.86	2.76	0.92
Chopped leaf	0.86	1.09	0.64	2.59	0.86
Whole root + stem	0.41	0.38	0.89	1.68	0.56
Whole leaf	0.11	0.2	0.2	0.51	0.17
Broccoli					
Macerated root + stem	0.32	0.15	0.36	0.83	0.28
Macerated leaf	0.74	0.36	0.3	1.4	0.47
Chopped root + stem	0.23	1.08	0.3	1.61	0.54
Chopped leaf	0.4	0.11	0.26	0.77	0.26
Whole root + stem	0.4	0.36	1.17	1.93	0.64
Whole leaf	0.41	0.3	0.56	1.27	0.42
Cauliflower					
Macerated root + stem	0.15	0.28	0.59	1.02	0.34
Macerated leaf	0.53	0.7	0.93	2.16	0.72
Chopped root + stem	0.15	0.11	0.3	0.56	0.19
Chopped leaf	0.28	0.56	0.61	1.45	0.48
Whole root + stem	1.15	1.29	1.21	3.65	1.22
Whole leaf	1.21	1.15	0.92	3.28	1.09
Cabbage cv. Scorpio					
Macerated root + stem	0.64	1.06	0.99	2.69	0.90
Macerated leaf	1.25	1.09	1.29	3.63	1.21
Chopped root + stem	0.89	0.36	0.52	1.77	0.59
Chopped leaf	0.91	0.4	0.26	1.57	0.52
Whole root + stem	0.08	0.41	0.3	0.79	0.26
Whole leaf	1.19	1.14	1.29	3.62	1.21
Cabbage cv. Rareball					
Macerated root + stem	1.24	1.28	1.23	3.75	1.25
Macerated leaf	0.64	0.97	0.56	2.17	0.72
Chopped root + stem	0.71	1.13	1.08	2.92	0.97
Chopped leaf	0.86	0.9	1.18	2.94	0.98
Whole root + stem	0.52	0.53	0.34	1.39	0.46
Whole leaf	1.3	1.45	0.96	3.71	1.24

Appendix Table 3**.** Continued...

TREATMENT	REPLICATION I	II	III	TOTAL	MEAN
Radish					
Macerated root + stem	0.81	0.76	0.72	2.29	0.76
Macerated leaf	0.83	0.6	0.6	2.03	0.68
Chopped root + stem	0.84	0.89	0.91	2.64	0.88
Chopped leaf	1.04	1.3	1.09	3.43	1.14
Whole root + stem	0.34	0.2	0.32	0.86	0.29
Whole leaf	0.56	0.38	0.3	1.24	0.41
Mustard					
Macerated root + stem	0.72	0.69	0.51	1.92	0.64
Macerated leaf	1.04	1.08	0.79	2.91	0.97
Chopped root + stem	0.74	1.23	1.1	3.07	1.02
Chopped leaf	0.28	0.18	0.67	1.13	0.38
Whole root + stem	0.82	0.9	0.34	2.06	0.69
Whole leaf	0.79	0.26	0.15	1.2	0.40
Pechay					
Macerated root + stem	0.74	0.8	0.9	2.44	0.81
Macerated leaf	1.24	1.06	1.3	3.6	1.20
Chopped root + stem	0.08	0.91	0.04	1.03	0.34
Chopped leaf	0.36	0.26	0.28	0.9	0.30
Whole root + stem	0.15	0.3	0.38	0.83	0.28
Whole leaf	0.87	0.51	0.36	1.74	0.58
Radish + mustard					
Macerated root + stem	0.23	0.56	0.4	1.19	0.40
Macerated leaf	0.15	0.04	0.2	0.39	0.13
Chopped root + stem	0.41	0.26	0.26	0.93	0.31
Chopped leaf	1.25	0.4	0.61	2.26	0.75
Whole root + stem	0.54	0.63	1.13	2.3	0.77
Whole leaf	1.21	0.97	0.66	2.84	0.95
Untreated	0.89	1.21	0.7	2.8	0.93
TOTAL	36.42	37.03	35.89	109.34	
MEAN	0.67	0.69	0.66		0.67

ANOVA TABLE

SOURCE OF VARIATION	DF	SS	MS	FC	Tab F 0.05	Tab F 0.01
Factor A	8	3.37	0.42	6.15**	2.03	2.69
Factor B	5	0.93	0.19	2.72*	2.3	3.2
A x B	40	12.37	0.31	4.52**	1.51	1.79
Error	108	7.39	0.07			
TOTAL	161	24.05				

CV = 38.18%

Appendix Table 4. Week three population of *Ralstonia solanacearum* in the soil (log cfu/g soil x 10^4)

TREATMENT	REPLICATION I	II	III	TOTAL	MEAN
Sunflower					
Macerated root + stem	0.38	0.4	0.38	1.16	0.39
Macerated leaf	0.53	0.75	0.43	1.71	0.57
Chopped root + stem	1.29	1.34	1.29	3.92	1.31
Chopped leaf	0.84	0.75	0.43	2.02	0.67
Whole root + stem	0.08	0.04	0.28	0.4	0.13
Whole leaf	0.58	1.04	0.08	1.7	0.57
Broccoli					
Macerated root + stem	0.61	0.86	0.7	2.17	0.72
Macerated leaf	0.41	0.45	0.41	1.27	0.42
Chopped root + stem	0.4	0.95	0.58	1.93	0.64
Chopped leaf	0.08	0.23	0.3	0.61	0.20
Whole root + stem	0.34	0.08	0.43	0.85	0.28
Whole leaf	0.34	0.15	0.08	0.57	0.19
Cauliflower					
Macerated root + stem	0.18	0.36	0.43	0.97	0.32
Macerated leaf	0.85	0.64	0.15	1.64	0.55
Chopped root + stem	0.26	0.6	1.05	1.91	0.64
Chopped leaf	0.94	0.94	0.59	2.47	0.82
Whole root + stem	0.71	0.92	0.75	2.38	0.79
Whole leaf	0.15	0.2	0.2	0.55	0.18
Cabbage cv. Scorpio					
Macerated root + stem	0.63	0.82	0.57	2.02	0.67
Macerated leaf	0.61	0.74	0.34	1.69	0.56
Chopped root + stem	0.4	0.64	0.43	1.47	0.49
Chopped leaf	0.97	1.31	0.95	3.23	1.08
Whole root + stem	0.08	0.15	0.15	0.38	0.13
Whole leaf	0.69	0.83	0.81	2.33	0.78
Cabbage cv. Rareball					
Macerated root + stem	0.87	0.85	0.95	2.67	0.89
Macerated leaf	0.3	0.23	0.53	1.06	0.35
Chopped root + stem	0.41	0.4	0.6	1.41	0.47
Chopped leaf	0.41	0.6	0.4	1.41	0.47
Whole root + stem	0.98	0.81	0.85	2.64	0.88
Whole leaf	0.99	0.91	0.96	2.86	0.95

Appendix Table 4. Continued...

TREATMENT	REPLICATION I	II	III	TOTAL	MEAN
Radish					
Macerated root + stem	0.95	0.92	0.98	2.85	0.95
Macerated leaf	0.11	0.26	0.23	0.6	0.20
Chopped root + stem	0.61	0.74	0.56	1.91	0.64
Chopped leaf	0.57	0.57	0.41	1.55	0.52
Whole root + stem	0.71	0.78	0.73	2.22	0.74
Whole leaf	0.36	0.32	0.32	1	0.33
Mustard					
Macerated root + stem	0.73	0.72	0.78	2.23	0.74
Macerated leaf	0.18	0.2	0.15	0.53	0.18
Chopped root + stem	0.77	0.85	0.88	2.5	0.83
Chopped leaf	0.23	0.15	0.87	1.25	0.42
Whole root + stem	0.28	0.64	0.72	1.64	0.55
Whole leaf	0.87	1.03	1.07	2.97	0.99
Pechay					
Macerated root + stem	0.58	0.81	0.8	2.19	0.73
Macerated leaf	0.43	0.56	1	1.99	0.66
Chopped root + stem	0.72	0.52	0.67	1.91	0.64
Chopped leaf	0.58	0.59	0.51	1.68	0.56
Whole root + stem	0.66	0.73	0.57	1.96	0.65
Whole leaf	0.78	0.67	0.62	2.07	0.69
Radish + mustard					
Macerated root + stem	0.26	0.28	0.2	0.74	0.25
Macerated leaf	0.58	0.69	1.07	2.34	0.78
Chopped root + stem	0.45	0.2	0.26	0.91	0.30
Chopped leaf	0.71	0.68	0.4	1.79	0.60
Whole root + stem	0.68	0.8	0.4	1.88	0.63
Whole leaf	0.58	0.7	0.72	2	0.67
Untreated	1.26	1.2	0.72	3.18	1.06
TOTAL	29.69	33.4	31.02	94.11	
MEAN	0.55	0.62	0.57		0.58

ANOVA TABLE

SOURCE OF VARIATION	DF	SS	MS	FC	Tab F 0.05	Tab F 0.01
Factor A	8	0.88	0.11	3.78**	2.03	2.69
Factor B	5	0.62	0.12	4.28**	2.3	3.2
A x B	40	9.19	0.23	7.91**	1.51	1.79
Error	108	3.14	0.03			
TOTAL	161	13.83				

CV = 29.35%

Appendix Table 5. Week four population of *Ralstonia solanacearum* in the soil (log cfu/g soil x 10^4)

TREATMENT	REPLICATION I	II	III	TOTAL	MEAN
Sunflower					
Macerated root + stem	0.48	0.73	0.81	2.02	0.67
Macerated leaf	0.4	0.28	0.58	1.26	0.42
Chopped root + stem	0.41	0.6	0.78	1.79	0.60
Chopped leaf	1.05	0.91	0.63	2.59	0.86
Whole root + stem	0.28	0.36	0.04	0.68	0.23
Whole leaf	0.8	0.36	0.26	1.42	0.47
Broccoli					
Macerated root + stem	0.26	0.34	0.3	0.9	0.30
Macerated leaf	0.26	0.2	0.2	0.66	0.22
Chopped root + stem	0.43	0.34	0.2	0.97	0.32
Chopped leaf	0.28	0.23	0.18	0.69	0.23
Whole root + stem	0.32	0.23	0.72	1.27	0.42
Whole leaf	0.45	0.54	0.32	1.31	0.44
Cauliflower					
Macerated root + stem	0.61	0.53	0.56	1.7	0.57
Macerated leaf	0.18	0.18	0.28	0.64	0.21
Chopped root + stem	0.36	0.49	0.79	1.64	0.55
Chopped leaf	0.48	0.08	0.11	0.67	0.22
Whole root + stem	0.38	0.43	0.48	1.29	0.43
Whole leaf	0.32	0.34	0.23	0.89	0.30
Cabbage cv. Scorpio					
Macerated root + stem	0.61	0.57	0.38	1.56	0.52
Macerated leaf	0.96	1.13	0.94	3.03	1.01
Chopped root + stem	0.9	0.78	0.53	2.21	0.74
Chopped leaf	0.08	0.08	0.26	0.42	0.14
Whole root + stem	0.2	0.82	0.65	1.67	0.56
Whole leaf	0.51	0.46	0.23	1.2	0.40
Cabbage cv. Rareball					
Macerated root + stem	0.58	0.71	0.63	1.92	0.64
Macerated leaf	0.41	0.23	0.38	1.02	0.34
Chopped root + stem	1.07	1.2	0.82	3.09	1.03
Chopped leaf	1	0.69	1.09	2.78	0.93
Whole root + stem	1.06	0.93	0.95	2.94	0.98
Whole leaf	0.04	0.18	0.2	0.42	0.14

Appendix Table 5. Continued...

TREATMENT	REPLICATION I	II	III	TOTAL	MEAN
Radish					
Macerated root + stem	0.28	0.6	0.54	1.42	0.47
Macerated leaf	0.86	0.96	1.18	3	1.00
Chopped root + stem	0.7	0.76	0.6	2.06	0.69
Chopped leaf	0.87	1.09	1.08	3.04	1.01
Whole root + stem	0.98	0.73	0.87	2.58	0.86
Whole leaf	0.6	0.71	0.36	1.67	0.56
Mustard					
Macerated root + stem	0.49	0.52	0.61	1.62	0.54
Macerated leaf	0.23	0.18	0.3	0.71	0.24
Chopped root + stem	0.41	0.34	0.76	1.51	0.50
Chopped leaf	1.02	0.89	0.97	2.88	0.96
Whole root + stem	0.97	0.84	1.06	2.87	0.96
Whole leaf	0.41	0.4	0	0.81	0.27
Pechay					
Macerated root + stem	0.43	0.94	0.85	2.22	0.74
Macerated leaf	0.7	0.96	0.86	2.52	0.84
Chopped root + stem	0.64	0.28	0.18	1.1	0.37
Chopped leaf	0.63	0.53	0.45	1.61	0.54
Whole root + stem	0.79	0.54	0.71	2.04	0.68
Whole leaf	0.72	0.99	0.79	2.5	0.83
Radish + mustard					
Macerated root + stem	0.32	0.36	0.18	0.86	0.29
Macerated leaf	0.61	0.51	0.89	2.01	0.67
Chopped root + stem	0.04	0.08	0.48	0.6	0.20
Chopped leaf	0.49	0.32	0.45	1.26	0.42
Whole root + stem	0.11	0.73	0.69	1.53	0.51
Whole leaf	0.57	0.51	0.46	1.54	0.51
Untreated	1.3	1.29	1.11	3.7	1.23
TOTAL	29.04	29.72	29.85	88.61	
MEAN	0.54	0.55	0.55		0.55

ANOVA TABLE

SOURCE OF VARIATION	DF	SS	MS	FC	Tab F 0.05	Tab F 0.01
Factor A	8	3.08	0.38	14.41**	2.03	2.69
Factor B	5	0.56	0.11	4.21**	2.3	3.2
A x B	40	7.27	0.18	6.81**	1.51	1.79
Error	108	2.89	0.03			
TOTAL	161	13.83				

CV = 29.88%

Appendix Table 6. Final population of *Ralstonia solanacearum* in the soil (log cfu/g soil x 10^4)

TREATMENT	REPLICATION I	II	III	TOTAL	MEAN
Sunflower					
Macerated root + stem	1.07	0.34	0.83	2.24	0.75
Macerated leaf	0.71	0.62	0.65	1.98	0.66
Chopped root + stem	1.09	1.08	1.1	3.27	1.09
Chopped leaf	0.26	0.23	0.34	0.83	0.28
Whole root + stem	0.08	0.23	0.18	0.49	0.16
Whole leaf	0.2	0.23	0.18	0.61	0.20
Broccoli					
Macerated root + stem	0.41	0.28	0.34	1.03	0.34
Macerated leaf	0.3	0.34	0.34	0.98	0.33
Chopped root + stem	0.41	0.2	0	0.61	0.20
Chopped leaf	0.15	0	0	0.15	0.05
Whole root + stem	0.43	0.51	0.41	1.35	0.45
Whole leaf	0.57	0.2	0.41	1.18	0.39
Cauliflower					
Macerated root + stem	0.66	0.51	0.49	1.66	0.55
Macerated leaf	0.32	0.41	0.34	1.07	0.36
Chopped root + stem	0.32	0.58	0.62	1.52	0.51
Chopped leaf	0.3	0.34	0.36	1	0.33
Whole root + stem	0.61	0.74	0.7	2.05	0.68
Whole leaf	0.18	0.08	0.3	0.56	0.19
Cabbage cv. Scorpio					
Macerated root + stem	0.04	0.08	0.11	0.23	0.08
Macerated leaf	0.87	0.96	0.76	2.59	0.86
Chopped root + stem	0.23	0.3	0.36	0.89	0.30
Chopped leaf	0.3	0	0	0.3	0.10
Whole root + stem	0.08	0.08	0.04	0.2	0.07
Whole leaf	0.11	0.45	1.01	1.57	0.52
Cabbage cv. Rareball					
Macerated root + stem	0.43	0.52	0.3	1.25	0.42
Macerated leaf	0.41	0.28	0.4	1.09	0.36
Chopped root + stem	0.96	0.46	0	1.42	0.47
Chopped leaf	0.28	0.26	0.23	0.77	0.26
Whole root + stem	0.83	1	0.81	2.64	0.88
Whole leaf	0.04	0.3	0.23	0.57	0.19

Appendix Table 6. Continued...

TREATMENT	REPLICATION I	II	III	TOTAL	MEAN
Radish					
Macerated root + stem	0.74	0.26	0	1	0.33
Macerated leaf	0.36	0.45	0.26	1.07	0.36
Chopped root + stem	0.66	0.89	0.95	2.5	0.83
Chopped leaf	0.91	0.99	1.03	2.93	0.98
Whole root + stem	0.78	0.51	0.43	1.72	0.57
Whole leaf	0.85	0.49	0.61	1.95	0.65
Mustard					
Macerated root + stem	0.79	0.71	0.66	2.16	0.72
Macerated leaf	0.08	0.26	0.23	0.57	0.19
Chopped root + stem	0.87	0.87	0.73	2.47	0.82
Chopped leaf	1.03	1	0.81	2.84	0.95
Whole root + stem	0.36	0.41	0.43	1.2	0.40
Whole leaf	0.71	0.61	1.15	2.47	0.82
Pechay					
Macerated root + stem	0.99	0.95	0.49	2.43	0.81
Macerated leaf	1.15	1.37	1.1	3.62	1.21
Chopped root + stem	0.18	0.18	0.15	0.51	0.17
Chopped leaf	0.4	0.36	0.28	1.04	0.35
Whole root + stem	0.3	0.92	0.72	1.94	0.65
Whole leaf	0.83	0.91	0.98	2.72	0.91
Radish + mustard					
Macerated root + stem	0.73	0.46	0.66	1.85	0.62
Macerated leaf	0.32	0.38	0.4	1.1	0.37
Chopped root + stem	0.58	0.51	0.53	1.62	0.54
Chopped leaf	0.2	0.08	0.23	0.51	0.17
Whole root + stem	0.7	0.8	0.54	2.04	0.68
Whole leaf	0.08	0.04	0.2	0.32	0.11
Untreated	1.43	1.3	1.21	3.94	1.31
TOTAL	27.25	26.02	25.41	78.68	
MEAN	0.50	0.51	0.47		0.49

ANOVA TABLE

SOURCE OF VARIATION	DF	SS	MS	FC	Tab F 0.05	Tab F 0.01
Factor A	8	2.86	0.36	13.31**	2.03	2.69
Factor B	5	0.50	0.10	3.73**	2.3	3.2
A x B	40	10.02	0.25	9.32**	1.51	1.79
Error	108	2.90	0.03			
TOTAL	161	16.29				

CV = 33.75%

Appendix Table 7. Changes in the population of *Ralstonia solanacearum* as affected by the combinations of biofumigant and tissue preparation (log cfu/g soil x 10^4).

TREATMENT	ASSESSMENT PERIOD					
	Initial	Week1	Week2	Week3	Week4	Week5
Sunflower						
Macerated root and stem	0.41^{lmnopq}	0.51^{klmno}	1.81^{abcd}	0.38^{ijklmnopqrs}	0.67^{cdefghi}	0.75^{cdefghij}
Macerated leaf	0.50^{hijklmnop}	1.24^{a}	0.71^{bcdefghijklm}	0.57^{efghijklmno}	0.42^{hijklmnop}	0.66^{cdefghijklm}
Chopped root and stem	0.86^{bcde}	0.93^{cdef}	0.92^{abcdefghij}	1.31^{a}	0.59^{efghijkl}	1.09^{ab}
Chopped leaf	0.66^{efghijkl}	0.81^{fgh}	0.86^{abcdefghijkl}	0.67^{cdefghijk}	0.8^{abcde}	0.27^{pqrstuvw}
Whole root and stem	0.44^{klmnop}	0.41^{opqrs}	0.56^{ghijklmnop}	0.13^{rs}	0.22^{nopq}	0.16^{stuvw}
Whole leaf	0.72^{defghijk}	0.63^{ijk}	0.77^{abcdefghijklmn}	0.57^{efghijklmno}	0.47^{hijklmnop}	0.20^{qrstuvw}
Broccoli						
Macerated root and stem	0.22^{pq}	0.3st	0.27^{nop}	0.72^{cdefghi}	0.30l^{mnopq}	0.34^{mnopqrstuvw}
Macerated leaf	0.63^{efghijklm}	0.65^{efg}	0.47^{hijklmnop}	0.42^{hijklmnopqrs}	0.22^{opq}	0.32^{nopqrstuvw}
Chopped root and stem	0.97^{bcd}	0.48^{mnop}	0.53^{ghjiklmnop}	0.64^{defghijklm}	0.32^{klmnopq}	0.20^{qrstuvw}
Chopped leaf	0.40^{lmnopq}	0.31^{rst}	0.25^{nop}	0.20^{pqrs}	0.23^{nopq}	0.5^{w}
Whole root and stem	0.59^{efghijklm}	0.82^{fgh}	0.64^{efghijklmnop}	0.28^{nopqrs}	0.42^{hijklmnopq}	0.45^{jklmnopqrs}
Whole leaf	0.85^{bcde}	0.34^{pqrst}	0.42^{ijklmnop}	0.19^{pqrs}	0.43^{hijklmnopq}	0.39l^{mnopqrstuv}

Appendix Table 7. Continued...

Cauliflower						
Macerated root and stem	0.51^{ghijklmnop}	0.55^{klmn}	0.34^{lmnop}	0.32^{lmnopqrs}	0.56^{efghijklm}	0.55^{ghijklmnop}
Macerated leaf	1.01^{abc}	0.61^{jklm}	0.72^{bcdefghijklm}	0.55^{efghijklmn}	0.21^{opq}	0.35l^{mnopqrtuvw}
Chopped root and stem	0.82^{bcdef}	0.34^{qrst}	0.18^{op}	0.63^{bcdef}	0.54^{efghijklmno}	0.51^{ijklmnopqr}
Chopped leaf	0.38l^{mnopq}	0.46^{nopq}	0.48^{hijklmnop}	0.82^{bcdefg}	0.22^{nopq}	0.33^{mnopqrstuvw}
Whole root and stem	0.30^{nopq}	0.70^{hij}	1.21^{ab}	0.79^{pqrs}	0.43^{hijklmnopq}	0.68^{cdefghijkl}
Whole leaf	1.07^{ab}	0.0.77^{gh}	1.09^{abcedf}	0.18^{pqrs}	0.29^{lmnop}	0.18^{rstuvw}
Cabbage cv. Scorpio						
Macerated root and stem	0.48^{jklmnop}	0.8^{efh}	0.89^{abcdefghjk}	0.67^{cdefghijk}	0.52^{fghijklmnop}	0.77^{uvw}
Macerated leaf	0.76^{cdefghjk}	1.03^{bc}	1.21^{abc}	0.56^{efghijklmno}	1.01^{a}	0.86^{bcdefg}
Chopped root and stem	0.80^{bcdefg}	0.90^{def}	0.59^{fghjiklmnop}	0.49^{fghijklmnopq}	0.73^{abcdefgh}	0.30^{opqrstuvw}
Chopped leaf	0.61^{efghijklm}	0.63^{ijk}	0.52^{ghijklmnop}	1.07^{ab}	0.14^{q}	0.10^{tuvw}
Whole root and stem	0.34^{mnoopq}	0.27^{t}	0.26^{nop}	0.13^{s}	0.56^{efghjiklm}	0.07^{vw}
Whole leaf	0.53^{ghijklmn}	0.24^{t}	1.2^{abc}	0.77^{bcdefg}	0.40^{ijklmnopq}	0.52^{hijklmnopq}
Cabbage cv. Rareball						
Macerated root and stem	0.54^{fghijklm}	1.12^{b}	1.25^{a}	0.89^{bcde}	0.64^{defghijk}	0.41^{klmnopqrst}
Macerated leaf	0.78^{cdefgh}	1.03^{bc}	0.72^{bcdefghijklmn}	0.35^{jklmnopqrs}	0.34^{jklmnopq}	0.36^{lmnopqrstuvw}

Appendix Table 7. Continued...

Chopped root and stem	046^{klmnopq}	0.49^{lmno}	0.97^{abcdefgh}	0.47^{ghijklmnopqr}	1.03^{a}	0.47^{jklmnopqrs}
Chopped leaf	0.24^{opq}	0.26^{t}	0.98^{abcdefgh}	0.47^{ghijklmnopqr}	0.92^{abcd}	0.25^{opqrstuvw}
Whole root and stem	0.28^{q}	0.24^{t}	0.46^{hijklmnop}	0.88^{bcde}	0.98^{abc}	0.88^{bcdef}
Whole leaf	0.2l^{mnopq}	0.28st	1.23^{ab}	0.95^{bcd}	0.14^{q}	0.19^{rstuvw}
Radish						
Macerated root and stem	0.38^{ijklmnopq}	0.23^{t}	0.76^{abcdefghijklmn}	0.95^{bcd}	0.47^{hijklmnop}	0.33^{mnopqrstuvw}
Macerated leaf	0.48^{fghijklmn}	0.41^{nopqrs}	0.68^{defghjijklmno}	0.20^{pqrs}	1.00^{ab}	0.35^{lmnopqrstuvw}
Chopped root and stem	0.54^{abc}	0.44^{nopq}	0.88^{abcdefghijk}	0.64^{defghijklm}	0.68^{bcdefghi}	0.83^{bcdefg}
Chopped leaf	1.05^{klmnopq}	1.04^{bc}	1.14^{abcde}	0.52^{fghijklmnop}	1.01^{a}	0.97^{abc}
Whole root and stem	0.47^{lmnopq}	0.45^{nopq}	0.28^{mnop}	0.74^{cdefgh}	0.86^{abcde}	0.57^{fghijklmn}
Whole leaf	0.43^{lmnopq}	0.52^{klmno}	0.41^{jklmnop}	0.33^{klmnopqrs}	0.55^{efghijklm}	0.65^{defghijklmn}
Mustard						
Macerated root and stem	1.25^{a}	1.29^{a}	0.64^{efghijklmnop}	0.74^{cdefgh}	0.54^{efghijklmno}	0.72^{cdefghijk}
Macerated leaf	0.87^{bcde}	0.33^{qrst}	0.97^{abcdefgh}	0.17^{qrs}	0.24^{mnopq}	0.19^{rstuvw}
Chopped root and stem	1.03^{abc}	0.85^{efg}	1.03^{abcdefg}	0.83^{bcdef}	0.50^{hijklmnop}	0.82^{bcdefghi}
Chopped leaf	0.59^{efghijklm}	0.75^{ghi}	0.37^{jlmnop}	0.42^{hijklmnopqrs}	0.96^{abcd}	0.94^{abcd}
Whole root and stem	1.02^{abc}	1.01^{bcd}	0.68^{cdefghijklmno}	0.54^{efghijklmno}	0.96^{abcd}	0.40^{klmnopqrstu}
Whole leaf	0.85^{bcde}	0.52^{klmno}	0.40^{jklmnop}	0.99^{bc}	0.27^{lmnopq}	0.82^{bcdefghi}

Appendix Table 7. Continued...

Pechay						
Macerated root and stem	0.61^{efghijklm}	0.95^{cde}	0.81^{abcdefghijklm}	0.73^{cdefghi}	0.74^{abcdefgh}	0.81^{bcdefghi}
Macerated leaf	0.46^{klmnopq}	0.43^{nopqr}	1.20^{abcd}	0.66^{cdefghijkl}	0.84^{abcdef}	1.21^{a}
Chopped root and stem	0.34^{mnopq}	0.27^{t}	0.34l^{mnop}	0.64^{defghijklm}	0.37^{ijklmnopq}	0.17^{stuvw}
Chopped leaf	0.47^{ghijklmno}	0.48^{mnop}	0.30^{mnop}	0.56^{efghijklmno}	0.54^{efghijklmno}	0.34^{mnopqrstuvw}
Whole root and stem	0.53^{cdefghi}	0.52^{klmnop}	0.27^{nop}	0.65^{cdefghijkl}	0.68^{bcdefghi}	0.64^{defghijklm}
Whole leaf	0.77^{cdefghi}	0.70^{hij}	0.58^{fghijklmnop}	0.69^{cdefghij}	0.83^{abcdefg}	0.90^{bcde}
Radish + mustard						
Macerated root and stem	0.44^{klmnopq}	0.29st	0.39^{jklmnop}	0.24^{opqrs}	0.29^{lmnopq}	0.61^{efghijklmno}
Macerated leaf	0.59^{efghijklm}	0.41^{opqrs}	0.13^{p}	0.78^{bcdefg}	0.67^{cdefghij}	0.36^{lmnopqrstuvw}
Chopped root and stem	0.97^{bcd}	1.05^{bc}	0.31^{mnop}	0.30^{mnopqrs}	0.20^{pq}	0.54^{ghijklmnop}
Chopped leaf	0.78^{cdefgh}	0.63^{ijk}	0.75^{abcdefghijklmn}	0.59^{efghijklmn}	0.42^{hijklmnopq}	0.17^{stuvw}
Whole root and stem	0.62^{efghijklm}	0.61^{jklm}	0.77^{abcdefghijklmn}	0.62^{defghijklm}	0.51^{ghijklmnop}	0.68^{cdefghijkl}
Whole leaf	0.65^{efghjikl}	0.61^{jkl}	0.94^{abcdefghi}	0.67^{cdefghijkl}	0.51^{fghijklmnop}	0.10^{tuvw}
Untreated	0.95^{a}	0.92^{a}	0.93^{a}	1.06^{a}	1.23^{a}	1.31^{a}