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**Research Article**

# Effect of Cultural and Biological Treatments in Managing Clubroot Disease of Cabbage in Sidhuwa, Nepal

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## Abstract

The *Plasmodiophora brassicae* Woronin is an important soil pathogen that attacks brassicaceae family of plants. The ability of pathogen to survive in soil for many years as a resting structure even in the absence of suitable host has increased its threat by making it more devastating. Lower effectiveness of treatments applied so far and failure to completely eradicate the disease once it enters the field has posed more problems. The study was conducted at a disease infected field in Sidhuwa, Dhankuta. It was focused on the effectiveness of different cultural and biological treatments against clubroot disease under field conditions. The treatments; lime, combination of lime and vermicompost, Effective Microorganisms (EM) solution and *Trichoderma viride* were arranged in Randomized Complete Block Design and replicated four times. The yield and yield attributing parameters and clubroot scale were recorded at the time of harvest. Values like Disease Incidence, Disease Severity Index and Disease Control Percentage were calculated based on clubroot scale value. The treatments showed no significant influence on vegetative parameters of the plants and on disease incidence. However, the combined treatment of lime and vermicompost was found to be most effective regarding disease severity and disease control i.e. the least Disease Severity Index (31.25%) and highest Disease Control Percentage (50.94%) compared to the control were obtained from the combined treatment of lime and vermicompost. Effective Microorganisms showed the highest disease severity. The combination of lime and vermicompost used as a treatment yielded highest marketable head weight (1.779 kg) and head diameter (19.90 cm).

**Keywords**: Clubroot; lime; vermicompost; *Trichoderma ;* EM; Severity

## Introduction

Vegetable crops are cultivated in only 7.3% of total cultivated land in Nepal (MoAD, 2010). *Brassica oleracea* cv. capitata (2n=18) is grown throughout Nepal as fresh cultivated vegetable (Rana, 1997). Cabbage contributes considerably in livelihood of even farmers with small land holding. Some of the areas with high potential of cabbage production are Kathmandu,Palung and Daman (Timila , 2007). Cabbage is produced in hills as well as terai and is one of the most important exportable vegetables of Nepal (Budathoki *et al.* , 2001).It can be cultivated in all types of fertile soils with good water regime (Chadha, 2001; Chatterjee, 1999). It does not grow well on a highly acidic soil but it is best grown above 6.5 pH (Singh *et al.* ,, 2004). It has high nutrient requirement, particularly nitrogen, phosphorous, boron and molybdenum in the soil (Mengel & kriby, 2004).Clubroot disease is an important disease affecting crucifer crops worldwide. It affects crops like

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cabbage, cauliflower, broccoli, Brussels sprout, radish, turnip, Broad leaved mustard, kale (Timila *et al.*, 2007). It is problematic in many districts like Bhaktapur, Lalitpur, Makwanpur, Dhankuta. (Timila, 2008). The production and area of cabbage in Dhankuta district was found to be reduced significantly from 39,417 mt and 1,505 ha in the year 2014/15 to 38,270 mt and 1,460 ha in the year 2015/16(MOAD,2015); (MOAD, 2016). Mono cropping of cabbage and cauliflower can increase disease incidence from 9 percent in the first year to 50 percent in the very next year causing 62 percent reduction in yield (Shrestha *et al.* , 1995). The presently adopted management strategies against the disease mainly compriseof the use of chemical pesticides which still have proven to be ineffective tackling the problem. The disease causes problems by forming galls or clubs on the roots such that roots get swollen and interferes with the uptake of water and nutrients (Gideon *et al.* , 2003). The invasion of plant root system by zoospores of *P. brassicae* is more favored by wet weather acidic condition (Kurowski, 2009). (Wallenhammar, 1996) suggested that its resting spores can persist in the soil for 20 years or more. Chemical pesticides create pollution, degrade environment, may be lethal to useful microorganisms, insects and may enter food chain by the process of eating and being eaten. Liming has been effectively used as control measure against clubroot disease since early 19$^{th}$ century (Dixon, 1996). Liming creates an unfavorable environment for clubroot pathogen most likely by disrupting the release of zoospores(Cassel, 1980). *Trichoderma spp* could be used as an effective biocontrol agent in controlling Clubroot disease (Timila, 2011). Cuevas *et al.* (2011) found out that, compared to conventional crop practices, the use of *Trichoderma* can reduce clubroot incidence by upto 45%. Organic amendments show positive effects in reducing incidence and severity of clubroot disease (Szczech *et al.*, 1993; Tilston *et al.*, 2002). Microbial activity in composts plays a major role for suppressing soilborne plant pathogens (Hoitink & Fahy, 1986). Effective microorganisms (EM) show high disease-suppressive potential with respect to agricultural and horticultural plants has been well documented (Kurowski *et al.*, 2009).

## Materials and Methods

The experiment was conducted at farmer's field at Chatthar-Jorpati rural municipality-6, Dhankuta district, Nepal from July 2017 to November 2017 at an elevation of around 2,175 meters from sea level. The field was susceptible to clubroot disease. Soil was sampled compositely before onset of research to assess soil pH. Clubroot susceptible Nepal Green-777 variety was used. The experiment was laid out in single factorial Randomized Complete Block Design with four replications and five treatments viz. Lime, combination of lime and vermicompost, Effective Microorganisms, *Trichoderma viride* and control. Individual plot area was dimensioned 1.2m* 1.2 m with 16 plants with a spacing of 40cm each between rows and between plants. The *Brassica oleracea* seedlings were raised on 2×1 m$^2$ area of nursery bed inside a plastic tunnel. Prior to transplanting, the main field was finely tilled and 10 days before transplanting, recommended dose of chemical fertilizers (240:180:80 kg NPK per ha) was applied to whole field. Lime was applied 2 weeks before transplating as per pH value of soil. Vermicompost (180g per transplanting hole) and *Trichodera viride* (5 lt/ha) were applied incorporated in soil before transplanting. Second dose of *Trichoderma* was soil drenched one month after transplanting. EM was soil drenched 1 week before transplanting at the rate of 1 lt/ha. Yield and yield attributing characteristics along with disease scoring were recorded at harvest. Fresh root weight was also recorded at harvest. Data tabulation was done with MS-Excel while analysis was done with GenStat (15$^{th}$ edition).

## Results and Discussion

### *Effect on Disease Incidence*

The effect of applied treatments on percentage incidence of clubroot disease was found to be non-significant. However the highest disease incidence (93.75%) was obtained for control treatment whereas least incidence (68.75%) was obtained for the treatment combination of lime and vermicompost followed by *Trichoderma* application (75%) (Table 1).

**Table 1:** Effect of different treatments on disease incidence

| Treatments | Percentage disease incidence | Percentage reduction in incidence compared to control |
|---|---|---|
| T1(control) | 93.75 (9.67)$^a$ | N/A |
| T2(lime) | 87.50 (9.33)$^a$ | 6.67 |
| T3(lime +vermicompost) | 68.75 (8.20)$^a$ | 26.67 |
| T4(Effective Microorganisms) | 87.50 (9.33)$^a$ | 6.67 |
| T5(*Trichoderma viride*) | 75.00 (8.66)$^a$ | 20 |
| F value | 0.225 (0.206)$^{ns}$ | |
| Grand mean | 82.5 (9.04) | |
| SEM+- | 7.99 (0.45) | |
| LSD | 24.61 (1.386) | |
| CV(%) | 19.4 (10) | |

Note: LSD,Least Significant Difference;SEM,Standard Error of Means;CV,Coefficient of Variation. Same letter(s) indicate the non significant difference between treatments based on LSD at 5% level of significance. Figures in parentheses represent square root transformed values.

The effectiveness of organic amendments like compost in reducing disease incidence was reported by many workers (Szczech *et al.*, 1993; Tilston *et al.*, 2002; Sabet, 2013). (Cuevas *et al.*, 2011) reported a reduction of clubroot disease incidence by 45% by the use of *Trichoderma*, as compared to fields with conventional crop practices. An experiment conducted at Bhaktapur suggested that lime reduced incidence by 33 percent (Timila, 2006). However this lower effectiveness of lime in present study might be due to different factors like difference in rainfall intensity causing differential leaching behavior of lime.

***Effect on Disease Severity Index (DSI) and Disease Control Percentage (DCP)***

Disease Severity Index value was significantly influenced by the applied treatments (Table 2). It was calculated based on a rating scale (0-3) given by (Kuginuki *et al.*, 1999) (modified by Xue et al, 2008). Severity value was ranged from 33.33% to 64.58%. It was highest (64.58%) for control followed by lime application (50%) whereas lowest(33.33%) for lime and compost combination. Treatments lime, EM, and *Trichoderma* were statistically at par. DCP compared to control was highest for the combination of lime and vermicompost (50.94%) and least for Effective Microorganisms (15.82%). Trembley *et al.* (2004) also supported the present result of lower effectiveness of lime over the combination of lime with compost by stating that the single use of lime often falls short of a satisfactory control of clubroot disease.Our findings were supported by (Szczech *et al.*, 1993; Tilston *et al.*, 2002). In a study conducted by (Davies & Jones, 2002) to test effectiveness of 9 organic amendments over control, compost showed good results in terms of reducing clubroot severity. For untreated plot, clubroot scale value was 5.9 (value ranging from 1-9) whereas it was 5.2 for composted plot. The present findings of highest Disease control for the treatment comprising compost was supported by (Noble, 2011) who found out in an experiment using 79 containers that when the soil was amended with at least 20% v/v compost, 59 containers showed suppressive effect against soil borne pathogens and only 6 showed disease promoting effect of compost amendment. However sufficient literatures on the use of vermicompost (as a specific form of compost) against clubroot disease were not found.


**Fig 1:** Rating of clubs of field plants

**Table 2** *Effect of different treatments on disease severityand disease control percentage*

| Treatments | Disease severity(%) | DCP compared to control | DCP compared to control |
|---|---|---|---|
| T1(control) | 64.58 (0.71) [a] | N/A | N/A |
| T2(lime) | 47.92 (0.50) [bc] | 26.54 (5.035) bc | 26.54 (5.035) bc |
| T3(lime +vermicompost) | 31.25 (0.31) [d] | 50.94 (7.066) a | 50.94 (7.066) a |
| T4(Effective Microorganisms) | 54.17 (0.57) [ab] | 15.82 (3.952) c | 15.82 (3.952) c |
| T5(*Trichoderma viride*) | 37.50 (0.39) [cd] | 42.26 (6.437) ab | 42.26 (6.437) ab |
| F value | <0.001*** | 0.025 (0.019) | 0.025 (0.019) |
| Grand mean | 47.1 (0.498) | 33.9 (5.62) | 33.9 (5.62) |
| SEM+- | 3.90 (0.0436) | 6.99 (0.593) | 6.99 (0.593) |
| LSD | 12.01 (0.134) | 22.35 (1.897) | 22.35 (1.897) |
| CV | 16.6 (17.5) | 41.2 (21.1) | 41.2 (21.1) |

Note: LSD,Least Significant Difference;SEM,Standard Error of Means;CV, Coefficient of Variation. Same letter(s) indicate the non significant difference between treatments based on LSD at 5% level of significance.Figures in parentheses of DSI column represent arcsine transformed values and of DCP column represent square root transformed values.

***Effect on Marketable Head Weight***

The treatments showed highly significant difference in head weight of cabbage. The treatment of combination of lime and vermicompost showed the highest head weight (1.779kg) followed by *Trichoderma* treatment (1.627kg) whereas control showed lowest head weight(1.175kg). The combination of lime and compost was statistically at par with *Trichoderma* treatment (Table 3).

In the present study, vermicompost incorporated treatment showed significantly higher head weight of cabbage compared to control. (Getnet & Raja, 2013) also showed that higher dose of vermicompost resulted in higher head weight in cabbage. (Jeyabal & Kuppusamy, 2001) showed that the integration of vermicompost, chemical fertilizer and biofertilizer increased yield in rice by 15.9% compared to chemical fertilizer alone. Improved nutrition due to organic amendments helped to improve soil physical condition and associated enhance in nutrient utilization (Lal & Kang, 1982).

***Effect on Head Diameter***

The equatorial head diameter of cabbage was significantly influenced by the treatments used. It was ranged from 19.90cm to 17.24cm. It was jointly highest (19.90cm) for the treatments consisting of lime and compost together, and *Trichoderma*. It was lowest for control treatment (17.24cm).The higher head diameter for the vermicompost combined with lime treatment was supported by (Getnet & Raja, 2013) who found out that among the vermicompost doses; 25g, 50g, 100g and 200g, highest dose resulted significantly higher head round measurement compared to other treatments and control. (Khatiwada *et al.*, 2011) also found a positive correlation between head weight and head diameter (Table 4).

**Table 3:** Effect of different treatments on head weight of cabbage

| Treatments | Head weight(kg) | Percentage increase in head weight(compared to control) |
|---|---|---|
| T1(control) | 1.175 [b] | 0 |
| T2(lime) | 1.296 [b] | 10.30 |
| T3(lime +vermicompost) | 1.779 [a] | 51.40 |
| T4(Effective Microorganisms) | 1.271 [b] | 8.17 |
| T5(*Trichoderma viride*) | 1.627 [a] | 38.47 |
| F value | 0.002 ** | |
| Grand mean | 1.430 | |
| SEM+- | 0.0928 | |
| LSD(0.05) | 0.2859 | |
| CV% | 13.0 | |

Note: LSD, Least Significant Difference; SEM, Standard Error of Means; CV, Coefficient of Variation. Same letter(s) indicate the non significant difference between treatments based on LSD at 5% level of significance.

**Table 4:** Effect of different treatments on head diameter of cabbage

| Treatments | Equatorial head diameter (cm) |
|---|---|
| T1(control) | 17.24 [c] |
| T2(lime) | 19.01 [ab] |
| T3(lime +vermicompost) | 19.90 [a] |
| T4(Effective Microorganisms) | 17.64 [bc] |
| T5(*Trichoderma* viride) | 19.90 [a] |
| F value | 0.048 |
| Grand mean | 18.74 |
| SEM+- | 0.688 |
| LSD(0.05) | 2.121 |
| CV% | 7.3 |

Note: LSD,Least Significant Difference;SEM,Standard Error of Means;CV,Coefficient of Variation. Same letter(s) indicate the non significant difference between treatments based on LSD at 5% level of significance.

***Effect on Fresh Root Weight***

The effect of treatments on root weight of cabbage during harvest was non-significant at 5% level of significance. It was ranged from 0.1550 kg to 0.1913 kg (Table 5). It was highest for EM treatment (0.1913kg) followed by control treatment(0.1844kg) and least for the treatment combining lime and vermicompost (0.1550kg). The severely affected roots showed higher weight due to increase in cell numbers and cell size. Primarily symptoms are seen in the root system the form of swellings and deformations known as clubs which are caused due to cellular hyperplasia (increase in cellular number) and cellular hypertrophy(increase in cellular size) (Hwang *et al.*, 2012)(Faggian & Strelkov, 2009) as cited in (Ramírez, 2016).

**Table 5:** Effect of different treatments on root weight of cabbage at harvest

| Treatments | Root weight(kg) |
|---|---|
| T1(control) | 0.1844 [ab] |
| T2(lime) | 0.1781 [ab] |
| T3(lime +vermicompost) | 0.1550 [b] |
| T4(Effective Microorganisms) | 0.1913 [a] |
| T5(*Trichoderma* viride) | 0.1563 [b] |
| F value | 0.074[ns] |
| Grand mean | 0.1730 |
| SEM+- | 0.00987 |
| LSD(0.05) | 0.03041 |
| CV% | 11.4 |

Note: LSD, Least Significant Difference; SEM, Standard Error of Means; CV, Coefficient of Variation. Same letter(s) indicate the non significant difference between treatments based on LSD at 5% level of significance

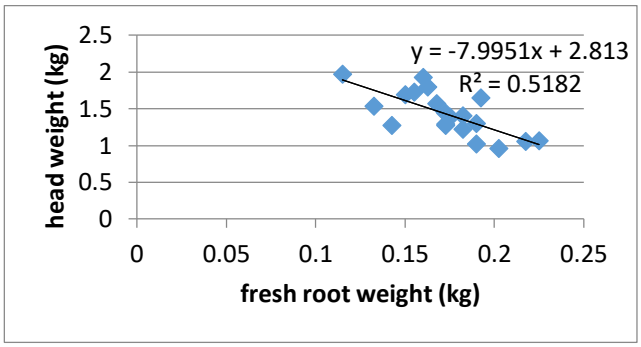


**Fig. 2:** Relationship between head weight and root weight of cabbage

A positive correlation was obtained between the severity of disease and root weight. Pasold & Ludwig-Muller (2013) also stated that smaller fresh root weight index (Ri/Rni) indicated less gall development leading to lower disease severity. However, Khatiwada *et al.* (2011) showed that with increase in clubroot severity, root weight decreased due to decaying of roots. In the case of present study, the decaying of roots might have been delayed by lower temperature than the experiment conducted by Khatiwada *et al.* (2011) at Palung and might be due to more moisture retention by hypertrophied roots. Drying might result to lowering of weight of roots.

## Conclusion

This study has highlighted possible treatments and farming practices that could be easily adopted by farmers to enhance clubroot control in the field. From the study, vermicompost and lime applied together was found to be highly effective against the disease in comparison to solo application of lime or EM application or *Trichoderma viride* application. The effect of treatments was non significant for all vegetative parameters. The least disease severity was observed for the combined treatment of lime and vermicompost (31.25 %) followed by *Trichoderma viride* (37.50 %). Highest head weight (1.779 kg) and head diameter(19.90cm) were also observed for the combined treatment of lime and vermicompost (1.779 kg). I advise farmers to adopt measures to improve soil qualities to take more yield by ultimately improving plant health. Incorporation of compost in addition to lime could be an easy yet useful method to improve soil health. The control of clubroot is complex and more research is still needed to discover more scientific mechanisms behind why treatments tested in this study controlled disease and how the soil and environmental factors in the field interacted with these treatments to alter their effectiveness across different sites.

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