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ECOFRIENDLY MANAGEMENT OF CERCOSPORA LEAF SPOT, CERCOSPORA COFFEICOLA (BERK & M.A. CURTIS) DISEASE OF COFFEE IN GULMI

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ABSTRACT
Cercospora disease is a very important disease of Coffee which results in yield loss and damage to the nursery plants in early stage of growth and later emerges a brown eye spot on cherries. Studies were carried out to find out ecofriendly way of disease management strategies against *Cercospora* leaf spot disease of Coffee using bio-rational treatments in the year 2021. The lowest mean disease incidence was recorded in Bordeaux mixture and Trichoderma treated block (34%). Bordeaux mixture was found superior in managing the disease with lowest PDI values (80%,57%,48%,44%) at respective four observation dates which was followed by *Trichoderma* (76%,67%,54%,46%) and Cow urine (63%,57%, 53%, 47%). Despite the significant superiority of Bordeaux mixture all the three treatments Bordeaux mixture, *Trichoderma* and cow urine were statistically at par with each other at last day of observation. Lowest mean AUDPC value and AUDPC value per day was recorded in block treated with Bordeaux mixture (2425.92 and 30.32) as compared to untreated check (4574.074 and 60.76) indicating the slower disease progress in Bordeaux mixture treated blocks followed by *Trichoderma* and cow urine (31.94 and 34.72). Despite of the superiority of Bordeaux mixture in the slow progress of disease all three treatments were statistically at par with each other. All the treatments were superior in controlling the disease over the control, among them Bordeaux mixture followed by Trichoderma is found to have superior efficacy in managing the disease.

KEY WORDS
Bordeaux, PDI, coffee, trichoderma, Nepal.

Agriculture, being the major source of livelihood of majority of the Nepalese people, contributes about 33% to the GDP and 65.6% to employment. Moreover, horticulture contributes about 14% to the total agriculture gross domestic product (World Economic Outlook Database, 2019).

Coffee is a high-value cash crop with important environmental implications that has been common among Nepalese for decades. It has spread to more than 40 districts in Nepal's middle hills. All Nepali coffee is of the Arabica variety, with a blend of bourbon and traditional varieties, and is cultivated by small farmers at altitudes ranging from 1000 meters to 1600 meters, using sustainable and environmentally friendly methods. Selective hand picking of fully ripe cherries is carried out and pulped immediately after harvesting with a mini hand pulper (wet processing), as well as any additional procedures that may be needed (Nationa Tea and Coffee Development Board, 2019). Coffee is cash crop started to be grown in Nepal almost with no use of inorganic fertilizer and pesticide (Agriculture Enterprise Centre, 2006). Coffee is cultivated mostly by resource-poor and small-scale farmers in Nepal's marginal highland areas. Chemical fertilizers and insecticides are seldom used in the production process. Highland and organic coffee, on the other hand, are well-known in worldwide markets because to their high-quality cupping and fragrance. Coffee contributes about 0.04 percent to GDP of Nepal (Postgraduate Student Society, 2004). It provides 5 times more yield than that of maize and millets and 2-3 times more yield than that of any other cash crops. Coffee being a perennial cash crop started to be grown in Nepal with almost no use of inorganic fertilizer and pesticide (Pandit, 2015).

Coffee was first brought in Nepal by Hira Giri from Myanmar back in 1938 in Aapachaur village, Gulmi. After that it remains curious plant for the several decades until his majesty's

government decided to import coffee seeds from India in 1968. Early 1980s considered the year for the significant progress in coffee farming with the development of Nepali Coffee Company & Coffee Development Centre under department of Agriculture in the same village (Coffee Promotion Program, 2014). Basic coffee growing districts of Nepal are Kaski, Palpa, Lamjung, Gorkha, Tanahun, Syanga, Baglung, Arghakhanchi, Parvat, Kavre, Lalitpur, Sindhupalchock The effective assessment of the plant disease to quantify the constraints to the production associated with plant disease plays a major role in the global food security, developing countries suffer more from plant diseases as compare to developed countries (Teng & W.C., 2001).Disease is the major constraints for quality coffee production. In Tropical America, Africa and Asia coffee is subjected to more than 40 diseases. These diseases are due to fungi, bacteria, virus, physiological disorders and mineral deficiencies. The disease being the major constraint for the organic coffee production. Tropical America, Africa and Asia are subjected to more than 40 coffee diseases due to various causes like fungi, bacteria, virus, physiological disorders and other mineral deficiencies (Gandia & Garcia, 1977).

Coffee is one of the most important cash generative crops in the mid hill regions of Nepal. Presently, coffee is cultivated in around 40 districts, but it has been producing commercially in about 20-22 hill districts (Sharma, Dhakal, Ghimire, & Rijal, 2015). Coffee was first introduced to Nepal in 1938 but it took a long time for commercial plantations to be established in the country. The first commercial plantation in Nepal was established in 1981, after which plantation growing spread to 41 districts in the country (Ranjitkar, et al., 2016). Coffee production in Nepal has increased from 72 tons in 2000 to 639 tons in 2019 growing at an average annual rate of 14.79%. In 2019, coffee production for Nepal was 639 tones, an increase of 13.78% compared to the previous year (Atlas, 2019).

Coffee diseases are caused by pathogenic and micro fungi and sometimes by bacteria and some viruses as well. The different parts of Coffee show disease symptoms causing debility, deformity and sometimes the whole plant die because of the disease (Waller J., 1985).*Cercospora* leaf spot, which is also known as brow eye spot disease in coffee is caused by fungal pathogen *Cercospora coffeicola.* It affects both leaves and fruits, which is seems more problematic in the newly established coffee orchard and is more susceptible to field with plant nutrition deficiency, particularly nitrogen and potassium and over-exposure to sunlight (Ribeyre & Avelino, 2012). *C. coffeicola* symptoms differ depending on which plant organ is infected. *Cercospora* "Leaf Spot" and *Cercospora* "Berry Blotch" are two of the disease's common names in which *Cercospora* is reference to the deuteromycete stage (Nelson, 2008). On the top leaf surface, lesions begin as chlorotic (yellow) patches that develop to become deep brown and necrotic (Virginia & Scot, 2015). Many of these spots have a yellow "halo" around the edges and a discolored, light center where sporulation might occur. *Cercospora* species generate the toxin cercosporin, which causes the halo (Nelson, 2008). Fruit symptoms typically appear 90 days after flowering (Gaitan, 2015) On green berries, this includes irregularly shaped brown, sunken lesions that are surrounded by a purple halo. Infected red cherries also have large, dark areas of sunken flesh. At this stage, fruit is susceptible to attack by opportunistic bacteria and fungi (such as *Colletotrichum gloeosporioides*), though symptoms from these organisms should not be falsely attributed to *Mycosphaerella.coffeicola* (Nelson, 2008).

MATERIALS AND METHODS OF RESEARCH

An experiment was conducted from Feb to June 2021 at the newly established orchard of Madane rural municipality 3 of Gulmi district with an elevation of 1300 masl. with longitude and latitude of 28° 11' 20" N and 83° 6' 17" E. The site was located on the North-West side of the Gulmi district, which lies in the range of tropical to temperate climate type and is emerging as suitable the site for coffee production in recent years. The location of the site with map of district is shown in the figure below (ArcGIS, n.d.).

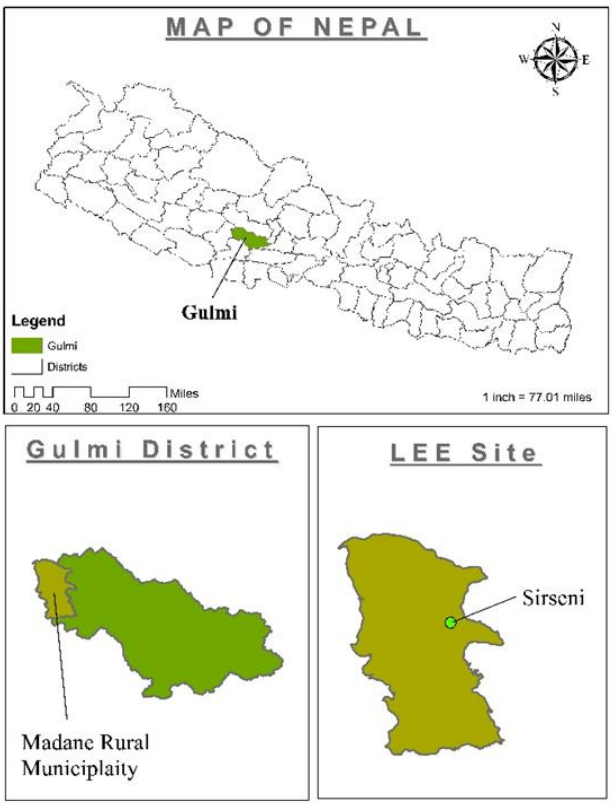

Figure 1 – Map showing Experimental Site

Experiment Design. Coffee being a perennial crop, sample was taken from the established orchard with standing crop of 1.5 yr. old. Three plants were considered as an experimental unit and were replicated three times. One piece of terrace/Kanla was considered as one replication. The design of the layout was conducted on Completely Randomized Block Design (RCBD). Blocking of the field was done after selecting the orchard affected with the *Cercospora* disease and each plant was selected randomly to be assessed. Further details of the treatments are shown below.

Table 1 – Details of Treatment used against Cercosopora Leaf spot disease of Coffee in Gulmi, Nepal

S.No.	Treatment's combination	Symbol	Concentration/Dose
1	Jholmol	T1	One parts in eight parts of water (100 ml in 800 ml water)
2	Trichoderma (Phytoderma 1.5% W.P.)	T2	5 gm per liter water
3	Neemcare (Neem oil 60%)	T3	5ml per liter water
4	Bordeaux mixture	T4	1:1:100 (Copper sulphate: Lime: Water)
5	Garlic extract	T5	10% (100 ml per one liter of water)
6	Cow urine	T6	One per eight parts of water (100 ml in 800 ml water)
7	Control	T7	Spraying of water

Application of Treatments. The amount of the treatments to be applied into the plants were calculated by using the general mathematical calculation under unitary method, where after final preparation of the treatments the volume of the treatments required for the plant to get completely wet were measured. The initial volume before spraying of the treatments was calculated by using the hand sprayer. The spraying was done so as to completely wet all the parts of the plants including stems and lower surface of the leaves. Bordeaux mixture, garlic extracts and cow urine were collected and prepared accordingly and were used for the day of treatments only so as to utilize its maximum use efficiency against the pathogenic fungus.

Weather Status of Experiment Site. The first few months of the field were not much cloudy and were sunny but at later stages the weather conditions fluctuate frequently with light to heavy rainfall occurring in the day and night time. Th details of the weather status of the experiment site are shown in the following charts (PN, 2021).

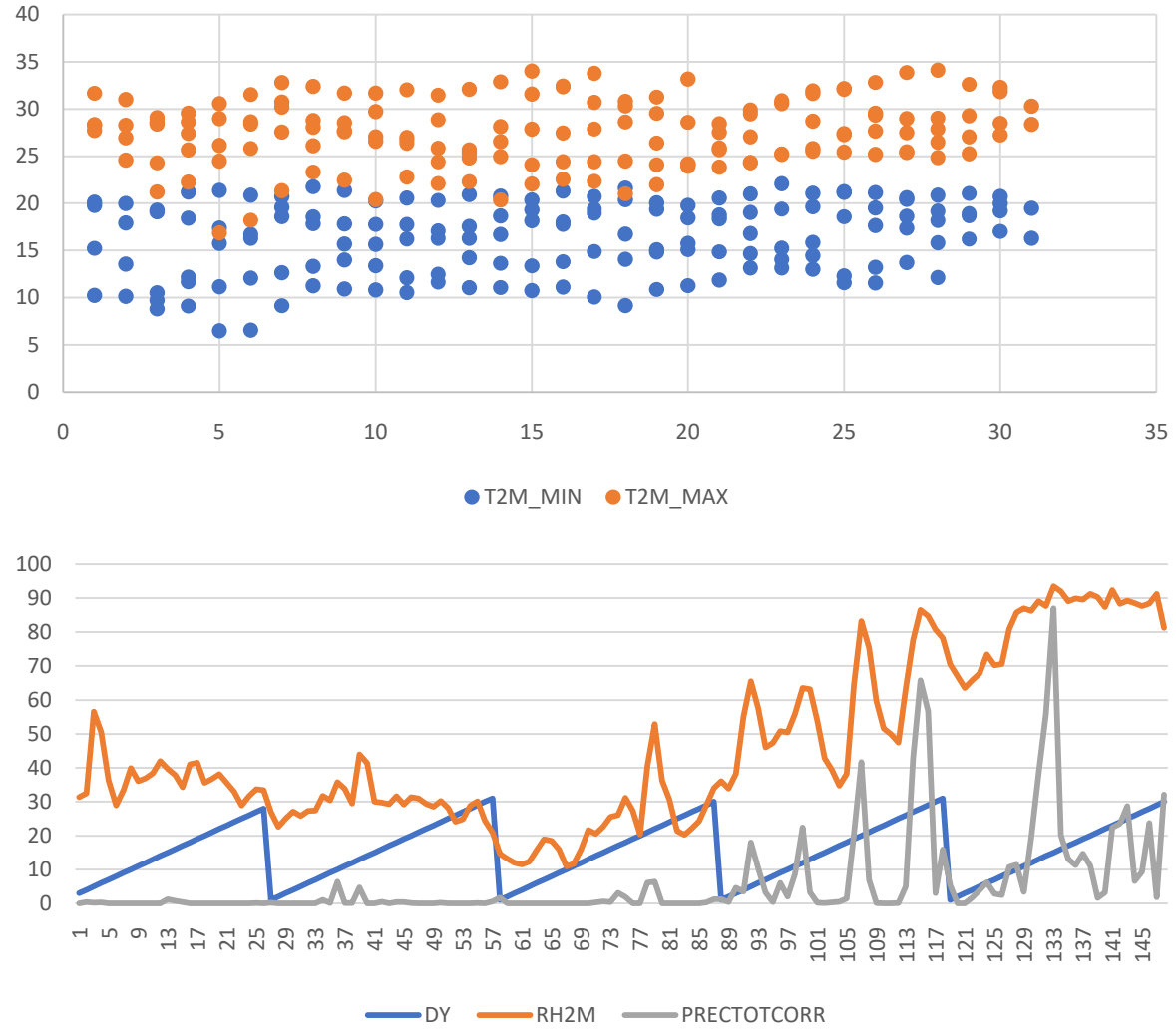

Figure 2 – Graph showing weather status of Experimental site (Source: https://power.larc.nasa.gov)

Pathological Observations:

A. Disease incidence: It is the percentage of diseased plants or parts in the sample or population of plants. It can be the proportion or percentage of diseased leaves in a plant, diseased stalks or a tiller or diseased seedlings in a field (Sharma P. N., 2017):

$$\text{Disease incidence} = \frac{\text{No .of infected units}}{\text{Total no.of units assessed}} \text{x}100$$

It can be generally applied to the disease in early stage and to those disease which affect the whole plants.

B. Disease severity: Disease severity is the percentage of relevant host tissues or organ covered by symptom or lesion or damaged by the disease. Severity results from the number and size of the lesions. DS is more appropriate in diseases like rusts, downy and powdery mildews, leaf spots and other similar disease. DS tells about the extent of damage caused by the disease. (Sharma P. N., 2017). It can be denoted by Percentage Disease Index/ Incidence and given by the formula:

$$\text{Disease Severity} = \frac{\text{Sum of total rating}}{\text{total number of observation} \times \text{highest grade in the scale}} \times 100$$

Estimation of area under disease progress curve (AUDPC). Different disease scores based on standard diagrammatic scale was used to calculate the AUDPC. The AUDPC could be calculated by using the following formula as used by (Das, Rajaram, Kronstad, Mundt, & Singh, 1993):

$$AUDPC = \sum_{i=1}^{n} (\mathrm{Yi} + 1 + \mathrm{Yi}) 0.5\ (\mathrm{Ti} + 1 - \mathrm{Ti})$$

Where: Yi = disease severity on the i^{th} date; Ti = date on which the disease was scored; n = numbers of dates on which disease was scored.

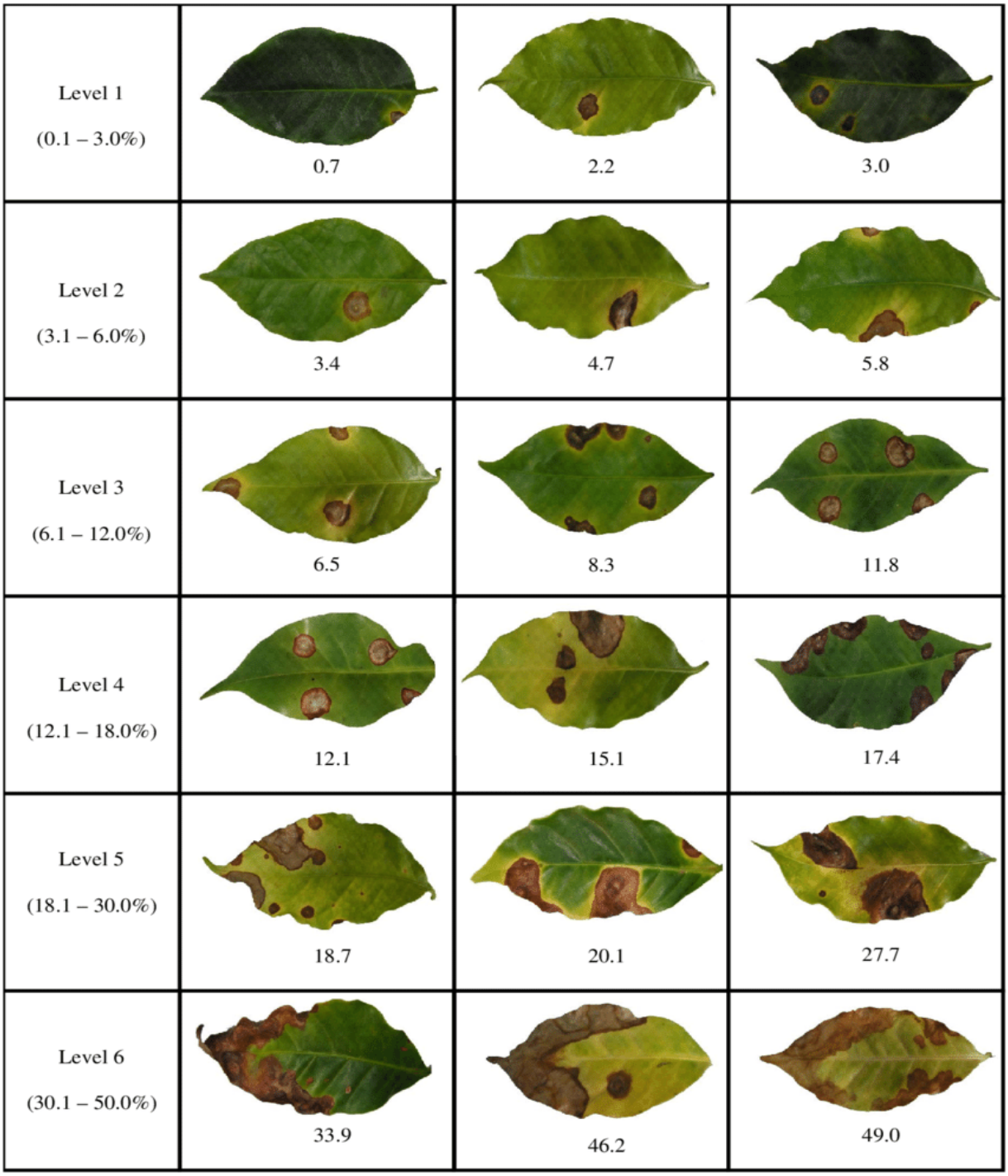

Figure 2 – Diagrammatic scales for Brown eye spot disease of Coffee

Different AUDPC values were calculated to know the disease progression over the different sprays time.

Efficiency on the basis of different treatments over the control was calculated by the following formula:

$$\text{Efficiency} = \frac{Treatment\ -ControlX\ 100}{Control}$$

Data recording. The recording parameters were leaf spot affected area (%), standard diagrammatic scale was used for the scoring of the disease. Disease incidence was recorded from each replication to find the efficacy of different control measures on disease. A diagrammatic scale with six levels (0.1-3.0; 3.1-6.0; 6.1-12.0; 12.1-18.0; 18.1-30.0; 30.1-49.0%) as developed by (Adriano Augusto et al., 2011) was used for the assessment of the disease.

Biometric Observations. Coffee being a perennial crop, the data was taken from the standing crop, three plants was taken as an experimental unit. Parameters to be studied to test the efficacy of different biocontrol management/ecofriendly measures are as follows:

- Percentage disease index (PDI)/ Disease Severity;
- AUDPC (Area Under Disease Progressive Curve);
- Disease Incidence;
- No. of infected leaves;
- Total no. of leaves.

The data was taken in 20 days interval, the scoring of the disease was done just before the spray of the treatments. The scoring of the disease was done by taking five leaves of same growth stage from the ground. The treatments were sprayed in equal interval in wind free and sunny hours of the day.

Statistical Analysis. Various data recorded on pathological and biometric parameters were arranged and compiled using Microsoft Excel 2010. The percentage data were transformed according to Arcsine transformation formula as and when required. Then, Data were subjected to ANOVA test by using R-3.6.2 and the significant treatment means were compared by using DMRT in accordance to Gomez and Gomez (1984). Means were compared by least significance difference (LSD) for treatment difference (Gomez & Gomez, 1984; Shrestha, 2019).

RESULTS OF STUDY

Effect of different treatments on disease incidence. The data presented in the given table revealed that highly significant differences for disease incidence among the treatments existed at 20 days after fourth treatment which means last treatment while no significant difference for the disease before spraying. All the management treatments were found superior in reducing the disease incidence as compared to untreated check at subsequent days after sprays. The lowest mean disease incidence as compared to untreated check after 20 days of first spray were found in block treated with *Trichoderma viridae* (51%) followed by cow urine (55%) which were statistically at par with other treatments Neem oil, Bordeaux mixture, garlic extract, Jholmol. Similarly, after four spray the mean disease incidence was found to be lowest in the block treated with Bordeaux mixture and *Trichoderma* (34%). After final observation all the treatments reduced the mean disease incidence and were statistically at par with each other. Whereas the lowest mean disease incidence was observed for the blocks treated with Bordeaux mixture and Trichoderma with disease incidence value of 34% in each block, which indicate that after final observation of the disease it was found to have a superior performance of Bordeaux mixture and Trichoderma over other treatments in reducing incidence of the disease. The reduction of the disease after the last days of observation in the untreated block might be due to the unfavorable conditions of the weathers and other factors which reduce disease incidence.

Table 1 – Effect of different treatments on disease severity of Cercospora leaf spot after four consecutive sprays at 20 days interval

	Mean Disease Incidence				
Treatments	Before spray	20 days after first spray	20 days after second spray	20 days after third spray	20 days after fourth spray
Jholmol	36.69 [a] (0.64)	30.51[b] (0.59)	17.98[b] (0.43)	16.66[b] (0.41)	14.14[b] (0.38)
Trichoderma	40.36 [a] (0.68)	24.41[b] (0.51)	21.79[b] (0.48)	14.28 [b] (0.38)	11.36[b] (0.34)
Neemcare	47.86 [a] (0.76)	32.43[b] (0.60)	26.51[b] (0.52)	19.49[b] (0.44)	16.18[b] (0.41)
Bordeaux mixture	47.57 [a] (0.76)	29.54[b] (0.57)	21.79[b] (0.48)	18.89[b] (0.44)	11.72[b] (0.34)
Garlic extract	43.71 [a] (0.71)	29.62[b] (0.57)	23.65[b] (0.50)	17.06[b] (0.42)	13.58[b] (0.37)
Cow urine	40.30 [a] (0.68)	28.11[b] (0.55)	21.24[b] (0.47)	16.65[b] (0.42)	14.73[b] (0.39)
Untreated	45.90 [a] (0.74)	49.83[a] (0.78)	48.77[a] (0.77)	40.99[a] (0.69)	39.60[a] (0.67)
SEM (±)	0.048	0.036	0.047	0.041	0.028
LSD(α=0.05)	0.388	0.112	0.145	0.127	0.087
CV	11.69	10.61	15.55	15.504	11.69
F test Probability Value	0.570	0.013	0.0056	0.0033	0.00030
Grand mean	43.20	31.67	25.96	20.57	17.33

Note: SEM: Standard error of mean, LSD: Least Significant Difference, CV: Coefficient of Variation, S: Significant, Values with same letters on columns are not significantly different at 5% DMRT (Duncan Multiple Range Test) and figure in parenthesis indicate arcsine transformed data.

Effect of different treatments on disease severity. The data presented in given table revealed that all the management treatments were significantly superior to the untreated check in reducing the disease severity at 20, 40, 60 and 80 days after first spray. Bordeaux mixture was found superior in managing the disease with lowest PDI values (80%,57%,48%,44%) at respective four observation dates which was followed by *Trichoderma* (76%,67%,54%,46%) and Cow urine (63%,57%, 53%, 47%). Despite the significant superiority of Bordeaux mixture all the three treatments Bordeaux mixture, *Trichoderma* and cow urine were statistically at par with each other at last day of observation.

However, the cow urine treated block found to have lowest disease severity (67%), which was superior among all the treatments applied and found to be more effective in terms of controlling disease in short term, which were statistically at par with Jholmol and *Trichoderma.* The use of Garlic extract, Bordeaux mixture and neem-based pesticides found to be inferior in early days of treatment application. Garlic extract also found to have an effective control over the cercospora disease which reduced the severity (PDI) from 88% to 57% from first day of spray to last day of observation, whereas Jholmol and Neemcare found to be statistically at par with Garlic extract, Bordeaux mixture and *Trichoderma.*

The value of PDI was found to be maximum for the control block with a value of 91% which were observed after final spray in other blocks, which indicate that all the treatments were superior in reducing the disease intensity of cercospora disease over control.

Effect of different treatments on AUDPC. The data presented in table revealed that the all the management treatments recorded relatively lower mean AUPDC values and AUPDC per day as compared to untreated check indicating the slower disease progress in treated blocks. Lowest mean AUDPC value and AUDPC value per day was recorded in block treated with Bordeaux mixture (2425.92 and 30.32) as compared to untreated check (4574.074 and 60.76) indicating the slower disease progress in Bordeaux mixture treated blocks followed by *Trichoderma* and cow urine (31.94 and 34.72). Despite of the superiority of Bordeaux mixture in the slow progress of disease all three treatments were statistically at par with each other. On the other hand, Jholmol, Neem based pesticide, and cow urine are also found to be significant which are statistically at par with *Trichoderma*, Bordeaux mixture in terms of AUDPC value (2888.88, 2925.926, 2777.77) and AUDPC per day value (36.11, 36.57, 34.72) indicating the somewhat slower progress of the disease.

Table 2 – Effect of different treatments on disease severity of Cercospora leaf spot after four consecutive sprays at 20 days interval

	PDI (Percentage Disease Index)				
Treatments	Before spray	20 Days after first spray	20 Days after second spray	20 Days after third spray	20 Days after fourth spray
Jholmol	48.14[a] (0.76)	38.88[bc] (0.67)	38.88 [b] (0.67)	31.47 [b] (0.80)	22.22 [bc] (0.49)
Trichoderma	46.29 [a] (0.74)	38.88[bc] (0.67)	27.77 [b] (0.54)	27.77 [b] (0.54)	20.36 [c] (0.46)
Neemcare	48.14 [a] (0.76)	48.14[abc] (0.76)	37.03 [b] (0.65)	25.92 [b] (0.53)	22.22 [bc] (0.49)
Bordeaux mixture	51.84 [a] (0.80)	51.84[ab] (0.80)	29.62 [b] (0.57)	22.21 [b] (0.48)	18.50 [c] (0.44)
Garlic extract	59.25 [a] (0.88)	53.70[ab] (0.82)	42.58 [b] (0.71)	35.18 [b] (0.63)	29.62[b] (0.57)
Cow urine	40.73 [a] (0.69)	35.18[c] (0.63)	29.62[b] (0.57)	25.92[b] (0.53)	20.36[c] (0.46)
Control	42.58 [a] (0.71)	59.25[a] (0.87)	61.10[a] (0.90)	55.55[a] (0.84)	62.96 [a] (0.91)
SEM (±)	0.062	0.047	0.069	0.0524	0.036
LSD(α=0.05)	0.1912	0.145	0.214	0.161	0.1002
CV	14.53	16.91	16.391	19.39	11.30
F test probability value	0.44915	0.022	0.04358	0.0082	2.969e-06
GM	48.14	46.55	38.08	32.005	28.038

Note: SEM: Standard error of mean, LSD: Least Significant Difference, CV: Coefficient of Variation, S: Significant, Values with same letters on columns are not significantly different at 5% DMRT (Duncan Multiple Range Test) and figure in parenthesis indicate arc sine transformed data.

Table 3 – Effect of different treatments on AUDPC values of Cercospora leaf spot of Coffee

Treatments	AUDPC	AUDPC per Day
Jholmol	2888.88[bc]	36.11[bc]
Trichoderma	2555.55[c]	31.94[c]
Neemcare	2925.926[bc]	36.57[bc]
Bordeaux mixture	2425.926[c]	30.32[c]
Garlic extract	3518.51[b]	43.98[b]
Cow Urine	2777.77[bc]	34.72[c]
Untreated	4574.074[a]	60.76[a]
SEM	255.95	3.199
LSD	731.43	9.14
CV	13.28	13.28
F test (α=0.05) probability	0.00051	0.0005107
GM	3095.23	38.69

Note: SEM: Standard error of mean, LSD: Least Significant Difference, CV: Coefficient of Variation, S: Significant, Values with same letters on columns are not significantly different at 5% DMRT (Duncan Multiple Range Test).

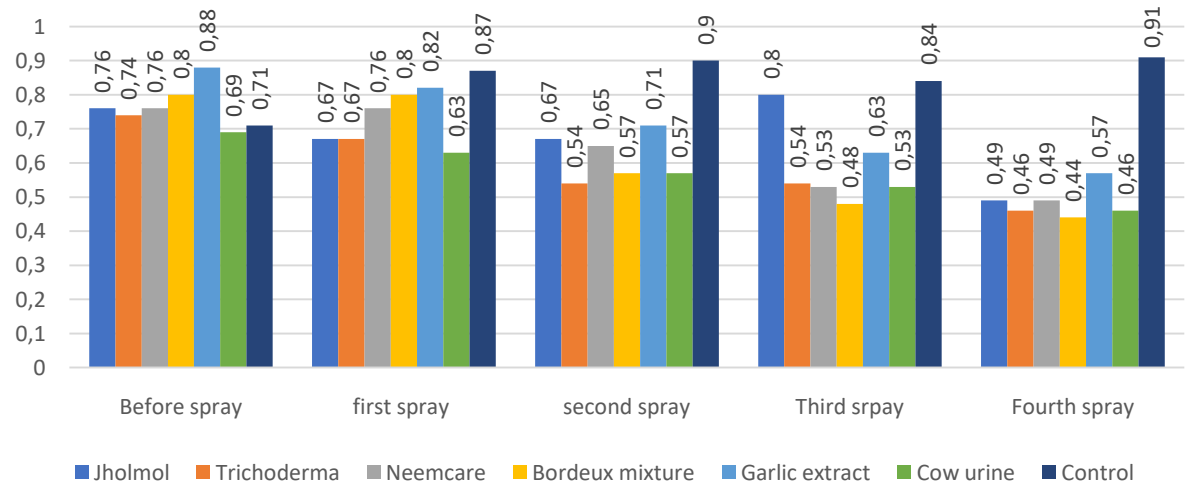

Figure 4 – Effect of different treatments on different AUDPC value over the period in Cercospora disease of coffee in Gulmi, 2021

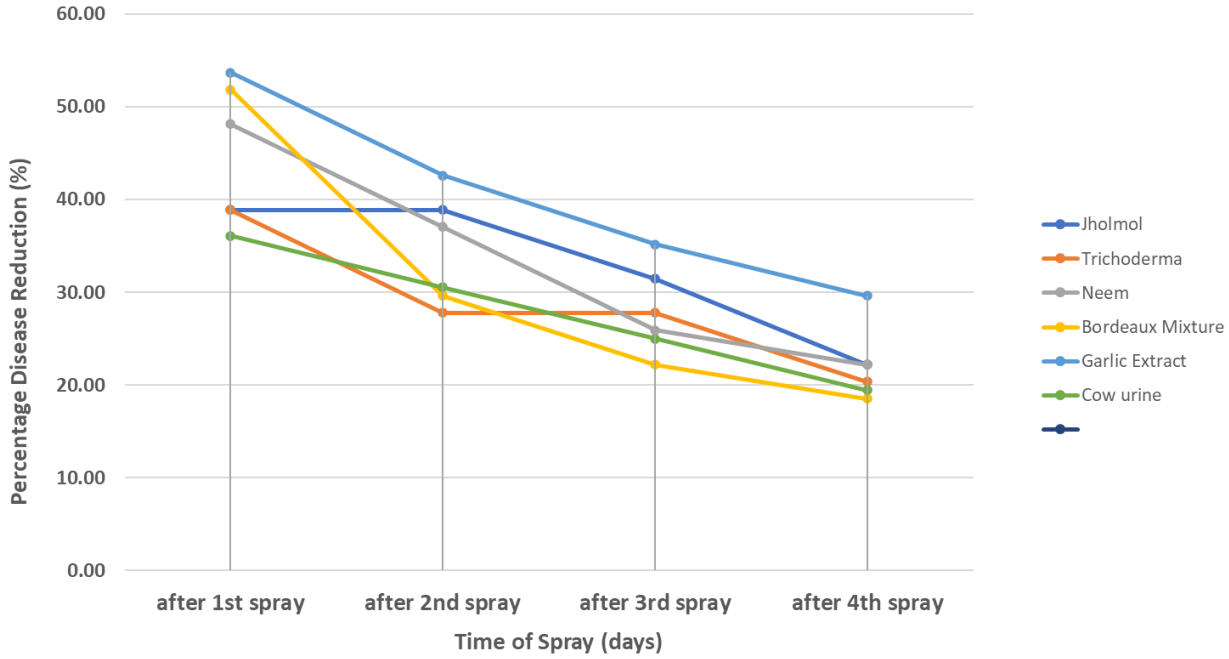

Figure 5 – Efficiency of different treatments over percentage disease reduction

Efficiency of different treatments over percentage disease reduction in cercopora disease. The percentage disease reduction was calculated using the formula given above to measure the efficiency of the different treatments over cercospora disease. From the graph it was found that Bordeaux mixture showed superior performance in reducing percentage of the disease in coffee. Which indicate that over other treatments Bordeaux mixture reduce the disease by greater extent from the value of 52% to 19%.

DISCUSSION OF RESULTS

The present work is done to disseminate simple, low cost & eco-friendly integrated package for this disease to farmers suitable under mid hill conditions. All the treatments significantly reduced the *Cercospora* disease severity after first, second, third and fourth sprays over unsprayed control. The result indicate that Bordeaux mixture is found to be must effective ecofriendly management treatments over other treatments which might be due to the toxicity of the copper to the pathogens as stated by (Agrios, 2005).

The efficacy of Bordeaux mixture is found to be superior over plant extracts in controlling fungal disease as stated by (Devika, Fatima & Bhimraj, 2017). Such effectiveness of Bordeaux mixture against pathogenic fungus was also reported by (Lee, Seo & Hwang, 2009). Management of the pathogenic fungus in medicinal plant omja showed a potent control efficacy of control values between 87 % and 96 %. Bordeaux mixture is found to be effective biological management for managing the fungus in the fruit trees due to its higher retention in the plant as reported by (Komarek, et al., 2009).Bordeaux mixture is found to reduce the one of the most destructive pathogenic fungal disease in grapevine disease incidence as compared to control (Romanazzi, et al., 2016).The findings of this study is also in accordance with the study done by (Mazaro, Mangnabosco, Citadin, Paulus, & Gouvea, 2013), where the lowest disease incidence occur which might be due to interference of the Bordeaux mixture with the *Mycosphaerella* pathogen in Strawberry.

The use of *Trichoderma* was also found to have antagonistic effect against cercospora lesion size and sporulation in sugar beet as stated by (Claudio, Cerato, Burzi, & Marinello, 2004). Similar findings were reported by (Hossain & Hosssain, 2013), (Zegeye, Santhanam, Gorfu, Tessera, & Kassa, 2011), (Mittal, 1996) under field conditions. Spray of Trichoderma in this study gave satisfactory result against *cercospora* leaf spot of okra (*Cercospora canscens*) over control with disease intensity of 20.20% (Bochalya, Lal, Simon, & Meena,

2017). The antagonistic effect of *Trichoderma* against *cercospora* pathogen might be due the nature of the genus *Trichoderma* in competing for nutrients and space, modifying the environmental condition, or promoting plant growth and plant defensive mechanisms and antibiosis or directly by mechanisms such as mycoparasitism (Raghavan & C.K., 2015). The use of Trichoderma for the control of cercospora was found to be best treatment to manage leaf spot (*Cersospora arachidicola* Hori) of Groundnut (Mapari & Zacharia, 2017). Many authors also recorded the antagonistic effect of Trichoderma sp. against pathogenic fungi. (Dennis & Webster, 1971) reported that Trichoderma spp. produced the antibiotic "Trichodermol". This antibiotic can inhibit the growth of several fungi. (Ahmed, 1995) tested the effect of some fungal filtrates on spore germination of S. fuliginea. He found that *T. harzianum* and *T. viride* had the most antagonistic effect on spore germination of the pathogen. (Sudha & Lakshmanan, 2007) reported that among the antagonistic microflora, maximum reduction in conidial germination was observed by *Trichoderma viride, T. Harzianum.*

(Jandaik, Thakur, & Kumar, 2015) reported that the cow urine has antifungal activities and the inhibitory activity can be used in the control of fungi. Also, the inhibitory activity of cow urine against fungal pathogens have been reported by (Rakesh, Dileep, Nawaz, Junaid, & Kekunda, 2013), (Jandaik, Thakur, & Kumar, 2015), (Katediya, Jaiman, & Acharya, 2019). Similar works had been conducted by (Chandel & Kumar, 2017) where among bio-formulations used garlic extract with cow urine and soap nut was found to be most effective with disease severity of 15.36% followed by cow urine with disease severity of 18.07%. In the study done by (Mishra, 2018) in fenugreek, foliar spray of cow urine minimized the disease intensity from 20.08 - 21.73% and it also increased the growth and yield of fenugreek. The cow urine was effective in reducing the sporulation in pathogenic fungus and cow urine can inhibit the sporulation, avoiding the spread of infection as reported by (Patil, et al., 2007).

The effect of plant extracts might be mainly due to the inhibitory effects of the antifungal compounds in the extracts on germination of the fungal spores, since some of these extracts in preventive treatment completely prevented infection which is also reported by (Daayf, Schmitt, & Belanger, 1995). All the plant extracts reduced the incidence and severity of cercospora leaf spot in both seasons compared to the untreated crops. However, neem seed and garlic clove extract significantly reduced the incidence and severity of the disease compared to the other plant extracts in the study done by (Bdliya & Alkali, 2010), in the same study the highest seed yield of 124 kg/ha and 3418 kg/ha was obtained by the use of cow urine. The lowest severity (PDI=4.55) was recorded in the *Cercospora* leaf spot of mungbean at 60 DAS.The use of plant extracts showed the reduction in disease incidence by about 16% and also the disease severity by 19% in *Cercospora* leaf spot of groundnut (S, G.R., K.N., K.N., & Kekuda, 2016).The inhibitory activity of garlic extract might be due to presence of presence of different compounds such as flavonoids, phenols, terpenes, alkaloids and others (Adriano Augusto, et al., 2011). Garlic extracts have been shown to contain at least several biologically active compounds and this diversity of compounds may account for the garlic extract's ability to affect a wide range of soilborne fungal pathogens (Avato, Tursi, Vitali, Candido, & Miccolis, 2000).

The Jholmol which contains fermented cow urine for about three weeks and more also contains fungicidal property as reported by (Malaco, et al.) against *Mycosphaerella fijiensis* causing black sigatoka of Banana (*Musa sp.)*.Similar fungicidal property of the unsterilized 15 days fermented cow urine was stated by (Katediya, Jaiman, & Acharya, 2019)., where they had found that the highest inhibition (68.2%) of the fungus against 20 percent concentration and 15 days fermented unsterilized cow urine.

CONCLUSION

Disease incidence was non-significant before the spray of the treatments, whereas found in block treated with *T. viridae* (51%) followed by cow urine (55%) which were statistically at par with other treatments Neem oil, Bordeaux mixture, garlic extract, Jholmol. Disease severity also found to have non-significant result before the spray of treatments,

whereas Bordeaux mixture was found superior in managing the disease with lowest PDI values (80%,57%,48%,44%) at respective four observation dates which was followed by *Trichoderma* (76%,67%,54%,46%) and Cow urine (63%,57%, 53%, 47%). Lowest mean AUDPC value and AUDPC value per day was recorded in block treated with Bordeaux mixture (2425.92 and 30.32) as compared to untreated check (4574.074 and 60.76) indicating the slower disease progress in Bordeaux mixture treated blocks followed by *Trichoderma* and cow urine (31.94 and 34.72). In overall Bordeaux mixture is found to have a greater efficacy in reducing the disease followed by Trichoderma and Cow urine. Other treatments also showed a superior performance over control in controlling the disease based on different pathological observations.

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CONFLICT OF INTERESTS

The authors declare that there is no conflict of interests regarding the publication of this paper.

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