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Effect of organic and inorganic soil amendments on bacterial wilt of tomato

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Abstract
Different organic and inorganic soil amendments *viz.*, Farmyard manure @15 t/ha, vermicompost @10 t/ha, poultry manure @6 t/ha, lime @0.5 t/ha, bleaching powder @0.015 t/ha and mustard cake @1 t/ha were evaluated to know their efficacy against percent plant mortality, population of *R. solanacearum* and soil pH. The data revealed that lowest percent plant mortality was observed in the treatment of bleaching powder @ 0.015 t/ha with 8% and 12% at 30 and 60 DAT, respectively with lowest population 7.6 and 23.2 cfu of *R. solanacearum* at 30 and 60 DAT, respectively. While the highest percent plant mortality was observed in control with 32% and 60% at 30 and 60 DAT, respectively with highest population 32.2 and 53.00 cfu of *R. solanacearum* at 30 and 60 DAT, respectively. Soil pH level was also significantly increased in the application of bleaching powder by 6.03 over the lowest pH was recorded in control with 5.65 pH at 30 DAT. While at 60 DAT, the highest pH was recorded in lime @ 0.5 t/ha with 6.2 over the lowest pH was recorded in control with 5.79. The coefficient of correlation were assessed between percent plant mortality, population of *R. solanacearum* and pH and found that the percent plant mortality and population of *R. solanacearum* were negatively correlated with pH that indicated increase in pH leads to decrease percent plant mortality and population of *R. solanacearum*.

Keywords: *R. solanacearum*, soil amendment, pH, bacterial wilt, tomato, *Solanum lycopersicum* L.

Introduction
Tomato (*Solanum lycopersicum* L.) is one of the most prevalent vegetable in the world due to its higher value of nutrition and versatile use. It causes heavy losses every year due to bacterial wilt disease caused by *Ralstonia solanacearum*. It is most destructive and devastating pathogen in warm temperate, tropical and sub-tropical areas of the world, leading to their heavy economic loss character the end of 19th century. This disease is more active mainly slightly acidic soils and diseases continuously occurred in such soils. Many workers have suggested several soil amendments according to soil pH for the management of bacterial wilt disease. The present study on the management of bacterial wilt has been carried out in the context of Chhattisgarh state.

The prevalence, survival, spread and occurrence of *R. solanacearum* is highly related to soil properties due to it's soil borne in nature. Moreover, the acidification of soil is strongly related to growth of *R. solanacearum*. Liming of soil is dominant and effective strategy to increase soil pH and reduce biotic and abiotic constraints which related to soil acidity. The use of organic fertilizer *viz.* vermicompost, farm yard manure, poultry manure and mustard cake is mostly recommended by several researchers in agriculture production for obtain plant nutrients and improve soils physical and biochemical properties (Chellemi and Marois, 1992; Brady and Weil, 2008 and van Elsas *et al.*, 2005) [3, 2, 17]. Organic soil amendments may leads to stimulating bio-control in soil therefore, the aim of present research work on efficacy of different doses of organic and inorganic soil amendments is to reduce the survival of *R. solanacearum* and increase the population of beneficial microorganism. To know the effectiveness of soil amendments, Lin *et al.* (2008) [11], Wu *et al.* (2014) and Ghosh *et al.* (2015) [6] evaluated various soil amendments and observed that the application of different soil amendments were suppressive against bacterial wilt disease of different solanaceous crops. Many workers worked to manage the bacterial wilt in different crop by neutralizing the pH of acidic soils (Dhital *et al.*, 1997; Chung *et al.*, 2004; Sharma *et al.*, 2010; He *et al.*, 2014; Li *et al.*, 2017; Xinshen *et al.*, 2020; Ajayasree *et al.*, 2021 and Tafesse *et al.*, 2021) [4, 5, 15, 20, 10, 16] and found that strong correlation between bacterial wilt incidence and soil pH. Yamazaki (2001) [21] observed the relationship between Ca nutrition in tomato and the activity of soil-born pathogen *R. solanacearum* in tomato field and reported reduced population of *R. solanacearum* and wilt severity when applied increased amount of Ca.

Materials and Methods

In the present study, the various organic and inorganic soil amendments were evaluated to know their efficacy against the population of *R. solanacearum* in soil, percent plant mortality of tomato plant and soil pH under a pot culture experiment during *rabi* 2021-22. The slightly acidic soil (pH 5.65) was mixed with the recommended dose of various organic and inorganic amendments *viz.*, farmyard manure @15 t/ha, vermicompost @10 t/ha, poultry manure @6 t/ha, lime @0.5 t/ha, bleaching powder @0.015 t/ha and mustard cake @1 t/ha and filled into the plastic pots. Seedling of tomato cultivar was prepared by standard nursery procedure and 25 days old healthy seedlings were transplanted into the plastic pots. Ten pots for each treatment with five plants were kept in each pot for experimental work.

Assessment of percent wilt incidence or percent plant mortality were assessed at 30 and 60 days after transplanting (DAT) by following formula (Horita and Tsuchiya, 2001) [8].

$$\text{\% wilt incidence or \% plant mortality} = \frac{\text{Number of wilted plant}}{\text{Total number of plant observed}} \times 100$$

For the assessment of *R. solanacearum*, population, the soil samples were collected from each pot of each treatment at 30 and 60 DAT and brought to the laboratory and prepared serial dilutions of each soil sample up to 10^{-7} dilution. Further, 25 micro liter of dilution 10^{-7} dilution for isolation of *R. solanacearum* was placed on triphenyl tetrazolium chloride (TZC) semi-selective media containing petriplates with the help of micropipette and spread well by spreader over the surface of media. Inoculated plates were incubated at 28 ^{0}C for 96-120 hours. Observations on colony forming unit (cfu) were recorded at 24 hours interval.

The soil pH was assessed with the help of a digital pH meter before potting of soil and at 30 and 60 DAT from each pot of each treatment. In this process, 10 g of soil from pot was mixed with 25 ml of distilled water in a beaker and mixed thoroughly with a stick and prepared solution. Then the pH value of all the samples was assessed by putting the electrode rod of the digital pH meter in the solution (Jackson, 1967) [9].

Result and Discussion

Percent plant mortality

It is clear from the obtained data presented in Table 1, Fig 1 and Plate 1, that the population of percent plant mortality caused by *R. solanacearum* was influenced in each treatment by the application of different inorganic and organic soil amendments. At 30 DAT under pot condition, soil application of bleaching powder @ 0.015 t/ha and was significantly reduced the percent plant mortality with 8% which was on par with the treatment of lime @ 0.5 t/ha with 12% followed by vermicompost @ 10 t/ha with 14%, FYM @ 15 t/ha and mustard cake @ 1 t/ha with each 18%, poultry manure @ 6 t/ha with 20% over control treatment (without any soil amendment) with 32% plant mortality observed. However at 60 DAT, soil application of bleaching powder @ 0.015 t/ha on soil were significantly reduced the percent plant mortality with 12% followed by lime @ 0.5 t/ha with 20%, vermicompost @ 10 t/ha with 22%, FYM @ 15 t/ha with 26%, poultry manure @ 6 t/ha with 34%, mustard cake @ 1 t/ha with 36% over the highest 56% plant mortality was recorded in control.

This was in agreement with the findings of Ghosh *et al.* (2015) [6] reported that application of bleaching powder @ 20 kg/ha followed by lime @ 500 kg/ha showed best result to reduce wilt disease incidence by 42.3% and 41.57% respectively. Similar findings reported by Ajayasree *et al.* (2021) [1] that lowest disease incidence as well as higher yield obtained in the combined application of bleaching powder @ 15 kg/ha + lime + streptocycline at 2g/10 lit + copper oxychloride @ 0.3%.

Population of *R. solanacearum*

It is clear from the obtained data presented in Table 1, Fig 2 and Plate 2, that the population of *R. solanacearum* was influenced in each treatment by the application of different inorganic and organic soil amendments. At 30 DAT under pot conditions, application of bleaching powder @ 0.015 t/ha was significantly reduced the population of *R.* solanacearum with 7.6 cfu followed by lime @ 0.5 t/ha with 12.00 cfu, vermicompost @ 10 t/ha with 12.2 cfu, FYM @ 15 t/ha with 16.2 cfu, poultry manure @ 6 t/ha with 21.8 cfu, mustard cake @ 1 t/ha with 27 cfu over the control recorded as maximum 32.2 cfu observed. At 60 DAT under pot conditions, application of bleaching powder @ 0.015 t/ha on soil significantly reduced the population of *R.* solanacearum 23.2 cfu followed by lime @ 0.5 t/ha with 29.00 cfu, vermicompost @ 10 t/ha with 22.4 cfu, FYM @ 15 t/ha with 29.20 cfu, mustard cake @ 1 t/ha with 33 cfu, poultry manure @ 6 t/ha with 36.4 cfu while control with maximum 53.00 colony forming unit observed.

The present result on application of lime was in agreement with the findings of Dhital *et al.* (1997) [5] to control *R. solanacearum* on potato that stable bleaching powder (SBP) @ 25 kg/ha reported the lowest bacterial population of *R. solanacearum* with 3.01 and 2.06 log cfu/g dry soil at 120 days at glasshouse as well as field conditions. Similar findings reported by Yamazaki (2001) [21] that concentration of Ca^{+2} ions and population of *R. solanacearun* was inversely proportional. When the concentration of Ca^{+2} ions increased then the population of *R. solanacearum* decreased. Phen *et al.* (2014) [14] reported the application of lime + urea mixture gave the best result by reducing the growth of *R. solanacearum* and control the disease rhizome rot of ginger.

Soil pH

It is clear from the obtained data presented in Table 1 and Fig 3, that the population of *R. solanacearum* was influenced in each treatment by the application of different inorganic and organic soil amendments. At 30 DAT under pot conditions, application of bleaching powder @ 0.015 t/ha on soil significantly increased the pH by 6.03 followed by lime @ 0.5 t/ha with 6.09, vermicompost @ 10 t/ha with 5.80, FYM @ 15 t/ha with 5.79, poultry manure and mustard cake at 6 t/ha and 1 t/ha respectively with each 5.77 pH, over control with no treatments 5.65 pH observed. At 60 DAT under pot conditions, application of lime @ 0.5 t/ha on soil significantly increased the pH by 6.21 which was on par with the treatments of bleaching powder @ 0.015 t/ha with 6.19 pH followed by vermicompost @ 10 t/ha with 5.93 pH, FYM @ 15 t/ha with 5.91 pH, poultry manure @ 6 t/ha with 5.86 pH, mustard cake @ 1 t/ha with 5.84 pH, over control with no treatments 5.79 pH observed.

In the present research work, the percent plant mortality was significantly decreased in each pot when different organic and inorganic soil amendments were applied in pot culture conditions. It may be due to increasing soil pH, soil health, beneficial microbial activity and decreasing *R. solanacearum* population. This was in agreement with the findings of Tafesse *et al.* (2021) [16] during researched in potato bacterial wilt in Ethiopia revealed that application of lime were significantly increases in soil pH where increase soil pH by p<0.001 may leads to decrease percent plant mortality by p<0.001, the more lime was applied increase pH significantly by strong positive effect. Similar findings by Li *et al.* (2017) [10], they conducted an experiment over 4 years in South China to record bacterial wilt disease, and observed that application of lime were significantly increase soil pH and reduces the incidence of wilt. These was in agreement by Viade *et al.* (2011) [18] reported that liming increases soil pH and improves soil condition in acidic soil due to increases alkaline earth metals and alkaline metals in soil, these features may be affected by the particle size of limestone where the finest lime particle in a single application yielded Ca and Mg concentrations in soils. Similar findings by Pang *et al.* (2019) [13] reported that application of lime was mitigate the acidity of soil and increases soil pH and beneficial microbes, the study compares the impact of lime application and without lime on microbial population, in comparison with no lime, lime increases soil pH and increases the level of base cations *viz.*, Ca^{+2} and Mg^{+2} and improve status of soil nutrients *viz.*, nitrogen and phosphorous as well as soil microbes *viz.*, endophytes, arbuscular mycorrhizae and ectomycorrhizae.

Correlation coefficient between percent plant mortality, population of *R. solanacearum* and soil pH

Correlation coefficient between percent plant mortality, population of *R. solanacearum* and soil pH on 30 and 60 DAT are presented in Table 2 (a and b).

Correlation coefficient between percent plant mortality, population of *R. solanacearum* and soil pH at 30 DAT were assessed and data are presented in Table 2 (a) that the percent plant mortality were positively correlated with population of *R. solanacearum* with 0.909288 value of correlation coefficient and percent plant mortality were negatively correlated with pH with -0.86973 value of correlation coefficient. Similarly, the populations of *R. solanacearum* were negatively correlated with pH with -0.83419 value of correlation coefficient. It indicated that increase in pH may lead to decrease in population of *R. solanacearum* and percent plant mortality with -0.83419 and -0.86973 value of correlation coefficient respectively. At 60 DAT, correlation coefficient between percent plant mortality, population of *R. solanacearum* and soil pH were presented in Table 2 (b) revealed that percent plant mortality were positively correlated with population of *R. solanacearum* with 0.946549 value of correlation coefficient and percent plant mortality were negatively correlated with pH with -0.80664 value of correlation coefficient. Similarly, populations of *R. solanacearum* were negatively correlated with pH with -0.85597 value of correlation coefficient. It indicates that increase in pH may lead to decrease in population of *R. solanacearum* and percent plant mortality with -0.85597 and -0.80664 value of correlation coefficient respectively.

The correlation coefficient data of present investigation was in agreement with findings of Tafesse *et al.* (2021) [16] reported that in field condition increase in pH by p<0.001 were decrease in percent plant mortality by p<0.001 during the study in potato bacterial wilt incidence at Ethiopia. They observed strongly correlation between percent plant mortality and soil pH. Similar findings reported by Li *et al.* (2017) [10], they conducted an experiment over 4 years in South China to record bacterial wilt disease and observed that there was strongly correlation between soil pH and percent plant mortality and application of lime were significantly increase soil pH and reduced the incidence of wilt. Similar findings by Michel and Mew (1997) [12] researched on soil amendments with urea and CaO on survival of bacterial wilt pathogen and reported that decrease in soil pH may leads to increases in percent plant mortality. Pathogen population decreases with a alkaline pH and resulted significantly lower pathogen population (p<0.001).

Table 1: Effect of different organic and inorganic soil amendments on percent plant mortality, population of *R. solanacearum* and soil pH

Treatments		Percent plant mortality		Population of *R. solanacearum*		Soil pH	
		30 DAT	60 DAT	30 DAT	60 DAT	30 DAT	60 DAT
T1	FYM @ 15 t/ha	18.00 (24.92)	26.00 (30.54)	16.20 (04.14)	36.00 (06.08)	05.79	05.91
T2	Vermicompost @ 10 t/ha	14.00 (21.67)	22.00 (27.88)	12.20 (03.63)	30.20 (05.59)	05.80	05.93
T3	Poultry @ 6 t/ha	20.00 (26.25)	34.00 (35.48)	21.80 (04.77)	47.80 (06.99)	05.77	05.86
T4	Lime @ 0.5 t/ha	12.00 (20.05)	20.00 (26.25)	12.00 (03.60)	29.00 (05.47)	06.03	06.21
T5	Bleaching powder @ 0.015 t/ha	08.00 (14.74)	12.00 (20.05)	07.60 (02.92)	23.20 (04.92)	06.09	06.19
T6	Mustard cake @ 1 t/ha	18.00 (24.92)	36.00 (36.80)	27.00 (05.28)	43.20 (06.65)	05.77	05.84
T7	Control	32.00 (34.27)	56.00 (48.44)	32.20 (05.76)	53.00 (07.35)	05.65	05.79
	SEm(±)	02.28	01.81	00.10	00.08	00.01	00.01
	C.D. (p=0.05)	06.64	05.28	00.28	00.23	00.03	00.04
	C.V. (%)	21.41	12.59	05.03	02.85	00.35	00.49

Values under parentheses are arcsine/angular transformed.

Table 2: Correlation coefficient between percent plant mortality, population of *R. solanacearum* and soil pH at 30 and 60 days after transplanting (DAT) of tomato

(a) Correlation coefficient between percent plant mortality, population of *R. solanacearum* and pH at 30 DAT

	Percent plant mortality	*R. solanacearum* population	Soil pH
Percent plant mortality	1	0.909288	-0.86973
R. solanacearum population	0.909288	1	-0.83419
pH	-0.86973	-0.83419	1

(b) Correlation coefficient between percent plant mortality, population of *R. solanacearum* and pH at 60 DAT

	Percent plant mortality	***R. solanacearum* population**	**Soil pH**
Percent plant mortality	1	0.946549	-0.80664
R. solanacearum population	0.946549	1	-0.85597
pH	-0.80664	-0.85597	1

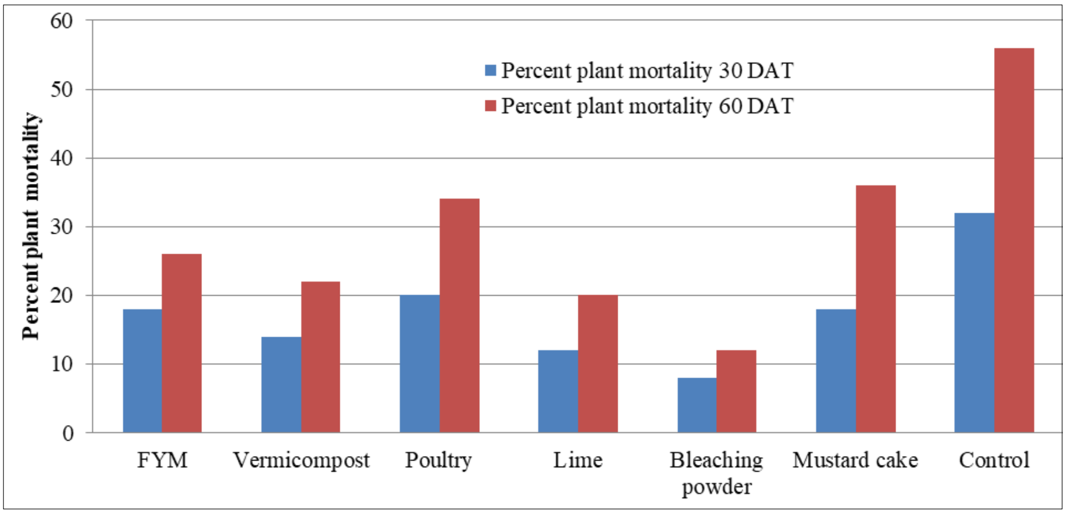

Fig 1: Effect of different organic and inorganic soil amendments on percent plant mortality

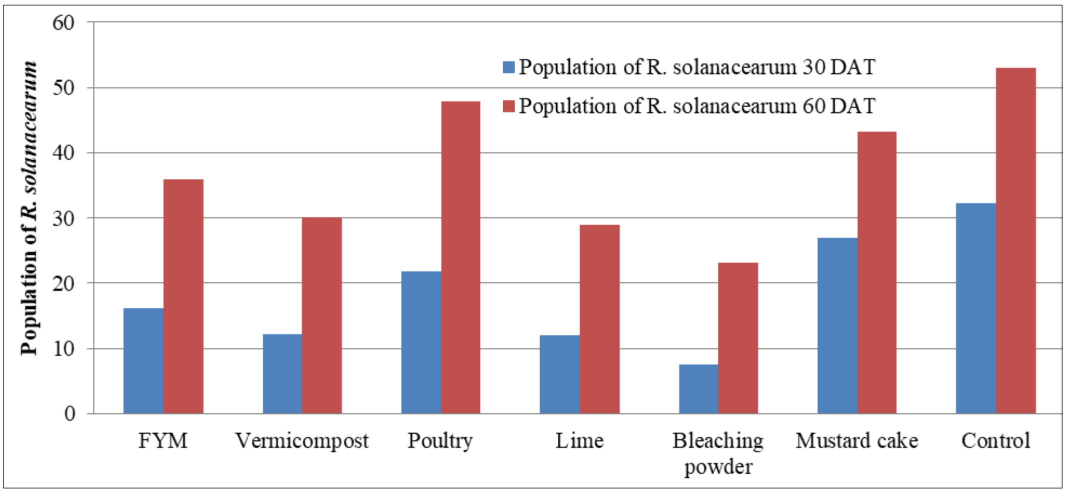

Fig 2: Effect of different organic and inorganic soil amendments on population of *R. solanacearum*

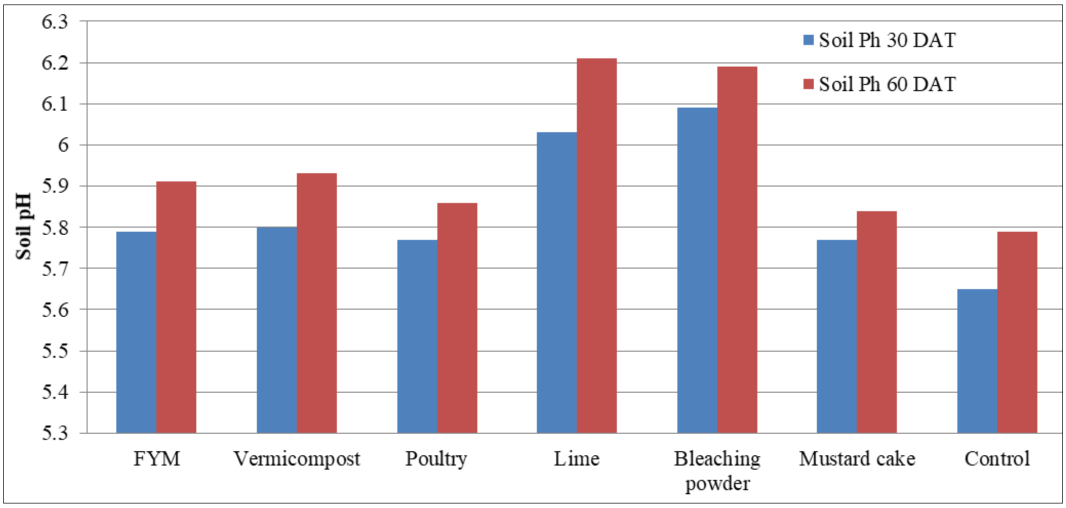

Fig 3: Effect of different organic and inorganic soil amendments on soil pH

Plate 1: Effect of different organic and inorganic soil amendments on percent plant mortality, population of *R. solanacearum* and soil Ph

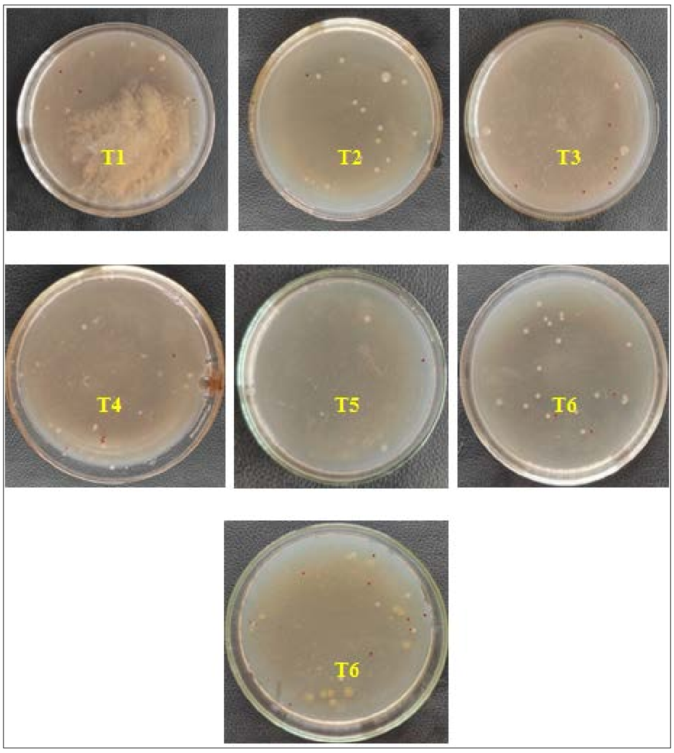

Plate 2: Effect of different organic and inorganic soil amendments on population of *R. solanacearum*

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