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Research Article

Influence of Soil Applied Plant Based and Synthetic Nitrification Inhibitors with Mineral Fertilizer on Microbial Populations under Cauliflower Cultivation

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ABSTRACT

The study was conducted to study the influence of soil applied plant derivatives of tree species combined with mineral fertilizer on microbial populations in cauliflower (Brassica oleracea var. Botrytis L). A field experiment was conducted during rabi season of 2018-19 and 2019-20, at experimental farm of University of Horticulture and Forestry, Nauni Solan. Eight treatment combinations viz. melia fruit powder, pomegranate rind powder, commercial neem cake, calcium carbide (CaC_2) @ 10 and 20 g kg^{-1} of soil, respectively and control were incorporated to study with three replications. Randomized block design was used to maintain heterogeneity. Results revealed that all the different treatments improved the microbial populations over the control treatment. Treatment of pomegranate rind powder@ 10 g kg^{-1} soil documented highest bacterial count (20.10 and 19.92 x 10^5 cfu g^{-1}soil), fungal count (4.48 and 4.43 x 10^3 cfu g^{-1}soil) and actinomycetes (3.45 and 3.39 x 10^3 cfu g^{-1}soil) during 2018-19 and 2019-20, respectively. However, the lowest count was recorded in control treatment. Moreover, melia fruit powder application also recorded higher microbial count in the soil as compared to synthetic inhibitor (CaC_2). Incorporation of natural inhibitors showed higher response and therefore sustained higher microbial populations under cauliflower.

Key words: *Nitrification inhibitors, Microbial population, Cauliflower, Sustainable agriculture.*

INTRODUCTION

The world's population has been projected to be around 9.6 billion by 2050 and for keeping pace with such tremendous growth in population, the agricultural production would also need to be increased by 70 per cent (Francesco & Mariangela, 2016). The increase in crop production can be expected either through promotion of improved high yielding cultivars or by increasing the cropping intensity.

Moreover, for sustainable production of agricultural and horticultural crops nutrient use efficiency would also have to be improved. The sub-optimal or supra-optimal dose of fertilizer applications especially of nitrogen fertilizers, leads to reduced nitrogen use efficiency, poor crop quality and ultimately results in lower yield levels. Application of fertilizers, especially nitrogen, more than the crop demands, leads to potential nitrate leaching from organic as well as inorganic N sources. In most of the intensive agricultural and horticultural production systems, around 50 per cent or even up to 75 per cent of the N applied to the field is not fully utilized by the plant and ultimately lost through leaching and volatilization from the soil nutrient pool.

Application of nitrogenous fertilizers has been considered as the major factor for increased agricultural productivity world-wide since last few decades. Among the different nutrients, nitrogen is regarded as a most important essential nutrient which controls the diversity and functioning of many terrestrial, freshwater and marine ecosystems. Nitrogen plays a major role in feeding the population, however huge application of nitrogenous fertilizers are being practiced in the developing countries causing hazards like land degradation. The agricultural production would have to be made sustainable over a long period of time, particularly in developing countries. Being a highly mobile and dynamic element nitrogen losses occurs through different ways i.e. NO_3^- leaching, denitrification, runoff, NH_3 volatilization, gaseous emissions of N_2O and NO to the atmosphere (Zaman et al., 2009). The losses from the fertilizers have long term adverse effects on the ecology and environment by means of eutrophication, losses in aquatic biodiversity and increased N_2O emissions (Warneke et al., 2011).

Nitrogen metabolism is primarily a microbially mediated process. Some microorganisms play an important role in improving the fertility of soil by metabolizing the N which is not taken up by the plants. However, it is a lengthy process and can cause

considerable risk of mineral N losses, as nitrate and urea are sparingly soluble in water and can be lost through run off or leaching in surface or sub- surface soils. Generally, cereal and vegetable based cropping systems are considered more prone to nitrogen (N) leaching losses. Adoption of proper management practices can lower these risks, but cannot fully solve it. Adoption of practices like crop rotation, mulching, zero tillage can also improve the N use efficiency, as the N left by one crop can be utilized by the succeeding crops.

Nitrification is a natural process occurring in the soil in which ammonium is converted into nitrite and further to nitrate form of nitrogen. The *Nitrosomonas* spp. of soil bacteria extract the energy from ammonium while converting it to nitrite form. Furthermore, another group of bacteria i.e. *Nitrobacter* spp. convert the nitrite form into nitrate. Both types of bacteria are commonly found in soil and determine the overall rate of nitrate production. Nitrification process occurs within few days or could take couple of weeks after application of ammonium sources, depending upon the soil temperature and moisture content.

Nitrification inhibitors are generally the compounds which restrict the *Nitrosomonas* bacteria activity and delay the nitrate production from ammonium. Adequate soil moisture conditions increase the activities of nitrification inhibitors as they are more favored to sandy soils with low organic matter and low temperatures. The use of nitrification inhibitors is of great significance as they maintain the higher amounts of inorganic nitrogen in its ammmonical form and thus results into lower leaching losses and other movements of N. Generally, there are several chemical and natural based inhibitors which have drawn special attention in last decade. The major inhibitors are 2-chloro-6-(trichloromethyl)-pyridine (Nitrapyrin), dicycandiamide (DCD), 3, 4-dimethylpyrazole phosphate (DMPP), CaC_2, neem cake and *Melia azedarach* (McCarty, 1999; Abbasi et al., 2003; Fangueiro et al., 2009; Khalil et al.,

2009; Zaman et al., 2009; Souri, 2010; & Pereira et al., 2010). Use of synthetic inhibitors have shown good edge of response over the natural one but use of these chemical inhibitors has been limited to only academic purpose as they bear higher cost of application, minimal availability and some side effects (Patra & Sukhma 2009). However, some plant based nitrification inhibitors also properties of nitrification inhibitions i.e. Karanji (*Pongamia glabra*), Neem (*Azadirachta indica*), Dharek (*Melia azedarach*. L) and tea (*Camellia sinensis)* have been reported by many researchers intheir studies (Freney et al., 2000, Kiran & Patra, 2003; Majumdar, 2002; Maureen et al., 2010; & Abbasi et al., 2011).

Cauliflower (*Brassica oleracea*) belongs to family Brassicaceae. It is an annual and cross pollinated plant which reproduces by seed. It is generally a low growing herbaceous plant. It grows into a long stalk with loosely arranged leaves and bear terminal curds which is also known as inflorescence. The stem is short and stout. In India, cauliflower occupies an area of about 469 thousand hectares with production of 9103 thousand metric tons per annum (Anonymous, 2019). In Himachal Pradesh, the crop occupies an area of about 5.56 thousand hectares with a production of 131.01 thousand tons per annum (Anonymous, 2018). Cultivation of cauliflower is done mainly on sandy to heavy soils rich in organic matter. Early crops prefer light soils while late crops grow better in heavier soils due to retention of moisture. A pH range of 5.5-6.5 is considered as optimum for growing cauliflower. In India, cauliflower is grown in large areas having a cool and moist climate. For a good seed germination of crop, a temperature of $10-21^0C$ is required. A temperature range of $15-21^0C$ is considered as optimum for growth and curd formation of the crop. Similarly, an optimum nutrition is required throughout its growing cycles making it imperative for proper nitrogen management. Very meager information is available on the use of these locally available plant derivatives as nitrification inhibitors and its impact on microbial populations on vegetable crops.

MATERIALS AND METHODS

A field experiments was conducted with eight treatment combinations replicated thrice in a randomized block design during *rabi* season 2018-19 and 2019-20 at the experimental farm of UHF Nauni, Solan (H.P.). Plot size of 6 m^2 (3m×2m) were selected and the one month old seedlings were used as transplanting material. Seedlings were sown at a planting distance of 60cm×45 cm for higher ease of handling in the raised beds to avoid water logging conditions. Recommended dose of NPK (127:76:72) were applied through sources viz. Urea, single super phosphate, murate of potash, respectively. FYM was applied as per university recommended dose (250 q ha^{-1}). All the different treatments were applied with required amount of fertilizers and FYM except absolute control. The plant based nitrification inhibitors were powdered and further applied as per the treatments. The following treatments were compelled in the study.

Table1. List of the experimental treatments

Serial No.	Treatments
T_1	Powdered pomegranate rind @10 g kg^{-1} of soil
T_2	Powdered pomegranate rind @20 g kg^{-1} of soil
T_3	Powdered melia fruits @10 g kg^{-1} of soil
T_4	Powdered melia fruits @20 g kg^{-1} of soil
T_5	Commercial neem cake @10 g Kg^{-1} of soil
T_6	Commercial neem cake @20 g kg^{-1} of soil
T_7	Calcium carbide (CaC_2) @ 15 g kg^{-1} of soil
T_8	Absolute control

RESULT AND DISCUSSION

Bacterial count

The application of plant derivatives had a pronounced effect on the bacterial population. An examination of the data presented in table (2) showed that during 2018-19 and 2019-20 the maximum bacterial count of 20.10×10^5 cfu g^{-1}soil and 19.92×10^5 cfu g^{-1}soil, respectively, were recorded in the treatment T_1 and the minimum respective bacterial counts of 15.93×10^5 cfu g^{-1} and 15.72×10^5 cfu g^{-1} soil were, however, recorded in control (T_8). The pooled data analysis again revealed a maximum bacterial count of 20.01×10^5 cfu g^{-1} soil in T_1 which was superior to all the other treatments and the minimum was in T_8 (control). Amongst the plant derivatives tested treatment pertaining to pomegranate rind @ 10 g kg^{-1} soil (T_1) significantly enhanced the bacterial population as compared to T_2, T_3 and T_4. The interaction T×Y showed that a maximum bacterial count of 20.10×10^5 cfu g^{-1} soil in T_1 during 2018-19 which was at par with the same treatment during 2019-20.

Fungal count

Table (2) clearly showed that the maximum fungal count of 4.48×10^3 cfu g^{-1}soil during 2018-19 was recorded in the treatment T_1 while the minimum fungal count of 2.79×10^3 cfu g^{-1} soil was in control (T_8). Similarly, in the year 2019-20 the maximum soil fungal count of 4.43×10^3 cfu g^{-1} soil was recorded in T_1 which was at par with T_5, while the minimum was again in control (T_8). The pooled data analysis revealed a maximum fungal count of 4.46×10^3 cfu g^{-1} soil in T_1 which was statistically superior to all other treatments. Amongst the new plant derivatives, melia fruit powder @ 10 g kg^{-1} (T_3) gave significantly higher fungal count of 4.30×10^3 cfu g^{-1} soil as compared to T_2, T_4 and T_7.

Table 2: Effect of different plant derivatives on microbial population of soil

Treatment *	Bacteria (x 10^5 cfu g^{-1}soil)			Fungi (x10^3 cfu g^{-1}soil))			Actinomycetes (x10^3 cfu g^{-1}soil))		
	2018-19	2019-20	Pooled	2018-19	2019-20	Pooled	2018-19	2019-20	Pooled
T_1: PR @ 10 g kg^{-1} soil	20.10	19.92	20.01	4.48	4.43	4.46	3.45	3.39	3.42
T_2: PR @ 20 g kg^{-1} soil	18.37	18.26	18.32	3.19	3.15	3.17	3.18	3.15	3.17
T_3: MF @ 10 g kg^{-1} soil	18.91	18.71	18.81	4.32	4.27	4.30	3.34	3.29	3.32
T_4: MF @ 20 g kg^{-1} soil	17.79	17.61	17.71	3.28	3.23	3.26	3.23	3.18	3.21
T_5: NC @ 10 g kg^{-1} soil	18.98	18.77	18.88	4.38	4.35	4.37	3.37	3.30	3.34
T_6: NC @ 20 g kg^{-1} soil	16.95	16.81	16.88	3.63	3.60	3.62	3.20	3.17	3.19
T_7: CaC_2 @15 g kg^{-1} soil	17.74	17.58	17.66	4.04	3.96	4.00	3.11	3.06	3.09
T_8: Absolute control	15.93	15.72	15.83	2.79	2.76	2.78	2.60	2.57	2.59
Mean	18.09	17.94		3.69	3.78		3.19	3.14	
CD (0.05)	1.44	2.80		0.02	0.08		0.02	0.09	
T		1.51			0.04			0.05	
Y		NS			NS			NS	
T × Y		2.14			0.06			0.07	

* RD of NPK were applied uniformly to all the treatments except T_8 (control)

PR = Pomegranate Rind   MF = Melia Fruits   NC = Neem Cake

Actinomycetes count

Data in table (2) revealed that during 2018-19 the maximum actinomycetes count (3.45×10^3 cfu g^{-1}soil) was recorded in the treatment T_1 comprising of the application of pomegranate rind @ 10 g kg^{-1} soil. The minimum actinomycetes count of 2.60×10^3 cfu g^{-1} soil was, however, recorded in control (T_8). Similar trend was also observed during the year 2019-20, with the maximum soil actinomycetes count of 3.39×10^3 cfu g^{-1}soil was recorded in T_1 which was at par with T_5 while the minimum was again in control (T_8). The pooled data analysis revealed a maximum actinomycetes count of 3.42×10^3 cfu g^{-1} soil in T_1 which was superior to all other treatments and minimum was in T_8 (control). The average actiomycetes count during 2018-

19 (3.19 x 10^3 cfu g^{-1} soil) was higher but not significantly as compared to that in 2019-20 (3.14 x 10^3 cfu g^{-1} soil). The interaction T×Y showed a maximum actinomycetes count of 3.45 x 10^3 cfu g^{-1} soil as in T_1 during 2018-19 was superior to 3.39 x 10^3 cfu g^{-1} soil in the same treatment during 2019-20.

In the present studies the application of nitrification inhibitors did not show a definite trend with respect to the nature of inhibitor used. The total microbial population varied with a maximum in Pomegranate rind @ 10 g kg^{-1} soil (T_1) but the other materials were also found to influence the microbial population controlling the biogeochemical cycles of the soils. The minimum microbial population (T_8) as the result of poor organic matter, nutrient and plant growth conditions in the absolute control.

The soil contains a dynamic population of microorganisms, which play an important role in the decomposition of organic matter, nitrogen fixation and transformation of elements. Combined application of nitrification inhibitors with chemicals might have reduced the activity of nitrifying bacteria, denitrifying bacteria, and net nitrification rates (Egamberdiyeva et al., 2001; & Patra & Sukhma, 2009). In another study it has been reported that the population of actinomycetes and bacteria recovered and then found to increase after 60 days of the application of Azotobacter sp. and nitrification inhibitor (*azadirachtin)* (Gopal et al., 2007). Many workers have reported that even application of nitrification inhibitors did not significantly affect the microbial population abundance and enzymatic activities (Maureen et al., 2010; Guo et al., 2019; Dong et al., 2013; & Xiuzhen et al., 2016). Maienza et al. (2014) reported that nitrification inhibitors addition significantly affected the microbial community structure and reduced fungal growth.

CONCLUSION

The study concluded that application of different plant based nitrification inhibitors had pronounced effect in the microbial populations over the control. Application of

pomegranate rind powder @ 10 g kg^{-1} soil improved the bacterial, fungal and actinomycetes populations over the rest treatments. Therefore, application of mineral fertilizers with pomegranate rind could be helpful and promising to sustain the microbial activities in soil under cauliflower cultivation in North Western Himalaya.

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