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Effect of natural farming, organic farming and integrated crop management practices on bacterial population, fungal population and dehydrogenase activity of soil at flowering stage of soybean + maize intercropping system

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Abstract
The present investigation entitled "Effect of natural farming, organic farming and integrated crop management practices on yield attributes and productivity of soybean + maize intercropping system under unbunded *Vertisol* of Chhattisgarh plains" was conducted during *Kharif* season of 2022 and 2023 at Instructional cum Research Farm, Indira Gandhi Krishi Vishwavidyalaya, Raipur, Chhattisgarh. The soil of experimental field was clayey (*Vertisol*), neutral in reaction (pH 6.85), medium in organic carbon (0.53%), low in available nitrogen (144 kg ha^{-1}), low in available phosphorus (18.4 kg ha^{-1}) and high in available potassium (377 kg ha^{-1}).

The experiment was carried out in randomized block design with five treatments and four replications. The treatments comprised of T_1 - Control (No addition of any inputs except labour for operations including weeding), T_2 - Complete Natural Farming (1. Beejamrit + Ghanjeevamrit + Jeevamrit; 2.Crop residue mulching; 3.Intercropping and 4.Whapasa), T_3 - AI-NPOF package (75% RDN through organic sources + two foliar spray of 10% cow urine and vermiwash at 30 and 50 DAS), T_4 - 50% RDN through organic sources + 50% RDN through inorganic sources (organic/natural pesticides for pest management) and T_5 - 50% RDN through organic sources + 50% RDN through inorganic sources (chemical pesticides for pest management).

Significantly the highest bacterial population (199.4, 214.6 and 207.0 cfu x 10^7 g^{-1} soil), fungal population (17.8, 19.4 and 18.6cfu x $10^4 g^{-1}$ soil) and dehydrogenase activity of soil (70.3, 71.7 and 71.0µg TPF g^{-1} soil) were found in AI-NPOF package (75% RDN through organic sources + two foliar spray of 10% cow urine and vermiwash at 30 and 50 DAS, respectively) during 2022, 2023 and on mean basis respectively followed by Complete Natural Farming (1. Beejamrit + Ghanjeevamrit + Jeevamrit; 2. Crop residue mulching; 3. Intercropping and 4. Whapasa).

Keywords: Natural farming, organic farming, Integrated crop management

Introduction
Natural farming, as the name implies, means that farmers do not need to purchase fertilizers and pesticides to ensure the growth of crops (Bishnoi *et al.*, 2017) [6]. Four pillars or components of natural farming are Beejamrit, Jeevamrit, Acchadana (Mulching) and Whaapasa (Moisture). Natural farming is a resource efficient farming system which minimizes the use of external resources and also restores the quality of soil and water resources. The importance of natural farming is to minimize the use of external inputs to the farm land and enrich soil through the propagation of soil microbes. It encourages the natural symbiosis of soil micro flora and crop plants. The natural inputs used in organic farming and natural farming are easily available, releases nutrients slowly, supplies macro and micro nutrients and provides favourable soil environment for microbial population and soil enzymes. General acceptance of organic farming and natural farming is not only due to the greater demand for pollution-free food but also due to natural advantage in supporting the sustainability in agriculture.

Though conventional farming helps in getting substantial yields, indiscriminate use of inorganic fertilizers and continuous farming has resulted in various soil hazards ultimately leading to lower productivity. Additionally, over emphasis on conventional farming has resulted in deterioration of soil and plant health. Restoring soil health by reverting to non-chemical agriculture has assumed great importance to attain sustainability in production. In this search for eco-friendly alternate systems of farming, organic and natural farming are increasingly becoming popular among the farming community with limited use of cow dung and cow urine (Patil *et al.,* 2022) [13].

For farmers with limited access to nutrient supplies, incorporating legumes into cereal-based cropping systems has long been recommended as a way to improve soil fertility and agro-ecological resilience (Snapp *et al.,* 1998; Thierfelder *et al*., 2012) [15, 18]. Cereal-legume based intercropping system is known to increase yield stability and is efficient at resource conservation and maintaining soil fertility. While agriculture directly contributes to 20% of greenhouse gas emissions in the country primarily due to livestock rearing and the use of nitrogenous fertilizers. These fertilizers are also the largest source of nitrate contamination in surface water bodies (Swaney *et al*., 2015) [17]. A new farming system came into light courtesy of Subhas Palekar Natural Farming System (SPNF) that is tailored fit for small and marginal farmer and Indian farmers that uses local indigents for farming like desi cow (*Bosindicus)* urine, cow dung, lime, gram flour and handful of soil and after fermentation it is used for foliar spray or fertigation. According to Subhas Palekar, natural farming components have high microbial load which upon application increase the soil flora that mineralize the soil macro and micro nutrients and make them available for plant use. Conjoint use of cereal-legume intercropping and natural farming systems can be ideal to reduce greenhouse gas emission and increase yield stability while maintaining soil fertility.

Globally, soils contain more carbon than plants and the atmosphere combined. Losing carbon-rich organic matter from soils releases carbon dioxide, a greenhouse gas, which can accelerate climate warming. But by regenerating soils, we can sequester more carbon underground and slow climate warming. In addition to protecting soil, cover crops take carbon out of the atmosphere as they grow and funnel it into the soil. Unlike cash crops that are harvested and removed from the soil, cover crops are left to decompose and contribute to soil formation. While plants are the source of carbon for soils, microbes control its fate by using it as food, thus ensuring that at least some of it will remain in the soil. Thus, it is believed that natural farming is based on the above hypothesis. With different interventions under it- adding microbes, adding cover crop, minimum tillage, multi-cropping, etc. it helps in soil regeneration and ultimately would lead to sustainable agricultural growth. There are several states practicing natural farming. Prominent among them are Andhra Pradesh, Chhattisgarh, Kerala, Gujarat, Himachal Pradesh, Jharkhand, Odisha, Madhya Pradesh, Rajasthan, Uttar Pradesh and Tamil Nadu. Till now 6.5 lakh ha. area is covered under natural farming in India (Anonymous, 2024) [2].

Adopting integrated nutrient management practices (organic manures, liquid manures with fertilizers) and certified organic agriculture (organic manures and biofertilizers) can reduce reliance on chemical inputs as well as make agriculture environmentally and economically sound. Organic farming is a production system that largely excludes or avoids the use of chemical fertilizers, pesticides, growth regulators, preservatives, livestock feed additives and totally rely on crop residues, animal manures, legumes, green manures, off-farm wastes, mechanical cultivation, mineral nutrient bearing rocks and biological pest control to maintain soil health, supply plant nutrients and minimize insects, weeds and other pests. Organic farming systems rely on the management of soil organic matter to enhance the chemical, biological and physical properties of the soil. Soil fertility management in organic systems depends on biologically-derived nutrient instead of using readily soluble forms of nutrients; less available forms of nutrients such as those in bulky organic materials. This requires the release of nutrients to the plant via the activity of soil microbes and soil animals. Apart from organic farming, another farming system called natural farming also involves similar components, which mainly depend on the use of naturally available inputs (Smith *et al.,* 2020) [14].

Integrated Crop Management (ICM) practices encompass a holistic approach to sustainable agriculture, combining traditional knowledge with modern technology to optimize resource use and improve crop yields. ICM involves a range of strategies such as crop rotation, intercropping, soil fertility management, and the use of organic and inorganic inputs judiciously. It emphasizes the importance of maintaining soil health through organic amendments and minimal tillage, conserving water through efficient irrigation systems, and enhancing biodiversity to naturally control pests and diseases. By integrating these diverse practices, ICM aims to increase productivity, reduce environmental impact, and support smallholder farmers by lowering input costs and promoting long-term agricultural sustainability.

Materials and Methods

The field experiment was conducted during *kharif*2022 and 2023 at the Instructional cum Research Farm, Indira Gandhi Krishi Vishwavidyalaya, Raipur, Chhattisgarh. The experiment was laid out in Randomized Block Design with four replications. The treatments comprised of five crop management practices. Soybean was taken as base crop and maize was taken as an intercrop in all the treatments with 4:2 row arrangement. The Soybean variety JS-9752 and maize variety RMH 4212 were taken in the experiment. The spacing used for soybean and maize was 30 cm x 7 cm and 60 cm x 20 cm, respectively. The sowing date of soybean was 5thJuly and 1st July and harvesting date was 21stOctober and 20th October during *kharif* 2022 and 2023, respectively. Whereas, the sowing date of maize was 5th July and 1st July and harvesting date was 3rdOctober and 29th September during *kharif*2022 and 2023, respectively. Prior to sowing, seeds were treated with Beejamrit @ 2.5 litres for 10kg seed in treatment 2 and with *Trichoderma* and *Pseudomonas* @ 5 g per kg seed in treatment 3, 4 and 5. The treatment details are presented in Table 1.

Table 1: Treatment details

Notation	Treatment
T_1	Control (No addition of any inputs except labour for operations including weeding)
T_2	Complete Natural Farming (1. Beejamrit + Ghanjeevamrit + Jeevamrit; 2. Crop residue mulching; 3. Intercropping and 4. Whapasa)
T_3	AI-NPOF package (75% RDN through organic sources + two foliar spray of 10% cow urine and vermiwash at 30 and 50 DAS)
T_4	50% RDN through organic sources + 50% RDN through inorganic sources (organic/natural pesticides for pest management)
T_5	50% RDN through organic sources + 50% RDN through inorganic sources (chemical pesticides for pest management)

Nutrient management was done as per the treatment. In case of AI-NPOF treatment 75% of recommended dose of nutrient was applied through organic sources *i.e.* 1/3rd FYM+ 1/3rd Vermicompost + 1/3rd Non-Edible Oil Cakes and two foliar spray of cow urine and vermiwash@ 10% at 30 and 50 DAS while in the treatments of integrated crop management 50% recommended dose of nutrients were applied through organic sources and rest of the 50% through inorganic sources.

RDN for soybean + maize - 25:60:30 kg ha^{-1} $N:P_2O_5:K_2O$ ha^{-1}

Soil microbiological properties

Determination of bacterial population in soil by serial dilution and pour plate method

Soil samples from the experimental field were collectedat flowering stage of crop. Population Count was recorded by the serial dilution and pour plate method. One gram of soil sample was suspended in 9 ml of sterilized water blanks. Ten folds of serial dilutions were prepared, from 10^{-1} to 10^{-7}, one ml of suspension from 10^{-6} to 10^{-7} fold Dilution was transferred in sterilized Petri plates. The sterilized nutrient agar medium at 45ºC temperature was poured into each Petri plate and mixed with an aliquot gently. After solidification of the medium, plates were kept at 30ºC temperature in an incubator for 4-5 days. The round, slimy bacterial colonies were observed on the medium. A number of colonies were counted with the help of a colony counter. The following formula was used to enumerate the bacterial population.

$$\text{No. of bacteria cfu /g. soil} = \frac{\text{Average plate count x dilution}}{\text{Weight of soil}}$$

Determination of fungal population in soil by serial dilution and pour plate method

Ten folds serial dilutions were prepared, from 10^{-1} to 10^{-7}. One ml of suspension from 10^{-4} and 10^{-5} fold dilution was transferred in sterilized Petri plates. The sterilized rose bengal medium at 45º C temperature was poured into each Petri plate and mixed with an aliquot gently. After solidification of the medium, plates were kept at 30 ºC temperature in an incubator for 5-6 days. Three replications were carried out for each dilution. The fungal colonies with cottony, fuzzy, hair like growth on the medium were counted under the colony counter. The fungal population per gram of soil was enumerated using the following formula.

$$\text{No. of fungi cfu /g. soil} = \frac{\text{Average plate count x dilution}}{\text{Weight of soil}}$$

Dehydrogenase activity (µg TPFh^{-1}g^{-1} soil)

The procedure to evaluate the dehydrogenase activity of soil was described by Klein *et al.* (1971) [10]. 1 g air dried soil sample from each treatment was taken in a 15 ml airtight screw capped test tube. 0.2 ml of 3% TTC solution was added in each of the tubes to saturate the soil. 0.5 ml of distilled water was also added in each tube. Gently tap the bottom of the tube to drive out all trapped oxygen so that a water seal was formed above the soil. No air bubbles were formed that was ensured. The tubes were incubated at 37 °C for 24 hrs. Then 10 ml of methanol was added. Shake it vigorously and allowed to stand for 6 hrs. Clear pink coloured supernatant was withdrawn and readings were taken by using UV/VIS Spectrophotometer with the wavelength of 485 nm and the dehydrogenase activity was worked out by the following formula:

Dehydrogenase activity (µg TPF h^{-1} g^{-1} soil) = (Reading of sample - Reading of blank) x 880

Result and Discussion

Bacterial population (cfu x 10^{7} g^{-1} soil)

The bacterial population was recorded at flowering stage of soybean + maize intercropping system (*kharif*season) and the data is presented in Table 2. During 2022, non-significant effect of the treatments was found on bacterial population.

Among the different treatments, AI-NPOF package (75% RDN through organic sources + two foliar spray of 10% cow urine and vermiwash at 30 and 50 DAS) recorded the highest bacterial population (214.6 and 207.0 cfu x 10^{7} g^{-1} soil) during 2023 and on mean basis, respectively which was comparable with rest of the treatments except control.

Fungal population (cfu x 10^{4} g^{-1} soil)

The fungal population was recorded at flowering stage of soybean + maize intercropping system (*kharif*season) and the data are presented in Table 2.

AI-NPOF package (75% RDN through organic sources + two foliar spray of 10% cow urine and vermiwash at 30 and 50 DAS) recorded the highest fungal population (17.8, 19.4 and 18.6 cfu x 10^{4} g^{-1} soil) which was statistically at par with Complete Natural Farming (1. Beejamrit + Ghanjeevamrit + Jeevamrit; 2. Crop residue mulching; 3. Intercropping and 4. Whapasa) during 2022, 2023 and on mean basis, respectively. The lowest fungal population was seen under control treatment.

Dehydrogenase activity (µg TPF g^{-1} soil)

Dehydrogenase activity was recorded at flowering stage of soybean + maize intercropping system (*kharif*season) and the data are presented in Table 2.

Among the different treatments, AI-NPOF package (75% RDN through organic sources + two foliar spray of 10% cow urine and vermiwash at 30 and 50 DAS) recorded the highest dehydrogenase activity (70.3, 71.7 and 71.0µg TPF g^{-1} soil) during 2022, 2023 and on mean basis, respectively and this was comparable with rest of the treatments except control which showed lowest dehydrogenase activity.

Discussion on microbiological properties of soil (*kharif* season)

AI-NPOF package (75% RDN through organic sources + two foliar spray of 10% cow urine and vermiwash at 30 and 50 DAS) recorded the highest bacterial population, fungal population and dehydrogenase activity followed by Complete Natural Farming (1. Beejamrit + Ghanjeevamrit + Jeevamrit; 2. Crop residue mulching; 3.Intercropping and 4.Whapasa).Soil micro-organisms play a very important role in soil fertility not only because of their ability to carry out bio-chemical transformation but also due to their importance as a source and sink for mineral nutrients. Use of organic manures is the object to accelerate microbial processes to enhance availability of nutrients in the assumable form. The microbial activities are enhanced as the fresh organic material acts as the nutrient source for the diverse soil flora and fauna. Organics modify the micro-climate, alter the environment of soil microbes, enhance soil flora and fauna activity, modify soil moisture regimes and properties associated with it and soil temperature in the root zone (Kiran *et al.* 2015) [9]. Cumulative effect of various sources of organic manures increases organic carbon content of soil which acts as energy source for microbes and their quick build up in the soil (Barik *et al*., 2006) [5]. Similar findings have been reported by Patil *et al.* (2023) [12] and Aher *et al.* (2015) [1].

Beejamrit, ghanjeevamrit and jeevamrit contain a lot of microbial properties and increase the soil micro flora with drastic increase in different soil enzymes. These formulations are rich in bacteria, fungus and actinomycetes population which not only provide basic soil conditioning but also have long lasting effect that leads to improvement in other soil biodiversity like soil arthropods, earthworms and other beneficiary fauna. Cow urine and dung based products are highly effective in improving the soil properties, and they increases the population of beneficial bacteria and fungus which act as antagonist against the plant pathogenic microorganisms (Gangadhar *et al.*, 2020) [8]. Devakumar *et al.* (2008) [7] and Sreenivasa *et al.* (2010) [16] have also reported the higher beneficial microbial population and the beneficial effect of jeevamrit in enhancing the microbial load in the soil. Barakzai *et al.* (2021) [4] and Vishwajeet (2020) also reported the same.

Dehydrogenase activity is strongly correlated with soil organic carbon content. Higher dehydrogenase activity in organic farming may be owing to higher organic matter content. The effect of organic sources on enzyme activities is probably a combined effect of a higher degree of stabilization of enzymes to humic substances and an increase in microbial biomass with increased soil carbon concentration Baishya *et al.* (2017) [3].

Table 2: Effect of natural farming, organic farming and integrated crop management practices on bacterial population, fungal population and dehydrogenase activity of soil at flowering stage of soybean + maize intercropping system

	Treatment	Bacterial population (cfu x 10^7 g^{-1} soil)			Fungal population (cfu x $10^4 g^{-1}$ soil)			Dehydrogenase activity (µg TPF g^{-1} soil)		
		2022	2023	Mean	2022	2023	Mean	2022	2023	Mean
T_1	Control (No addition of any inputs except labour for operations including weeding)	172.2	168.5	170.3	11.4	10.8	11.1	58.7	55.8	57.2
T_2	Complete Natural Farming (1. Beejamrit + Ghanjeevamrit + Jeevamrit; 2. Crop residue mulching; 3. Intercropping and 4. Whapasa)	195.6	208.3	202.0	16.3	18.8	17.5	66.8	68.9	67.9
T_3	AI-NPOF package (75% RDN through organic sources + two foliar spray of 10% cow urine and vermiwash at 30 and 50 DAS)	199.4	214.6	207.0	17.8	19.4	18.6	70.3	71.7	71.0
T_4	50% RDN through organic sources + 50% RDN through inorganic sources (organic/natural pesticides for pest management)	186.4	194.4	190.4	15.2	16.8	16.0	64.1	65.1	64.6
T_5	50% RDN through organic sources + 50% RDN through inorganic sources (chemical pesticides for pest management)	183.5	189.8	186.6	14.6	16.2	15.4	63.6	64.6	64.1
	SEm±	9.36	9.75	9.55	0.75	0.82	0.79	3.11	2.94	3.02
	CD (P=0.05)	NS	30.03	29.44	2.32	2.53	2.42	9.58	9.06	9.31

Conclusion

- The highest bacterial population was observed under AI-NPOF package (75% RDN through organic sources + two foliar spray of 10% cow urine and vermiwash at 30 and 50 DAS) which was comparable with rest of the treatments except control during 2023 and on mean basis.
- The highest fungal population was recorded under AI-NPOF package (75% RDN through organic sources + two foliar spray of 10% cow urine and vermiwash at 30 and 50 DAS) which was statistically at par with Complete Natural Farming (1. Beejamrit + Ghanjeevamrit + Jeevamrit; 2. Crop residue mulching; 3. Intercropping and 4. Whapasa).
- The highest dehydrogenase activity was recorded under AI-NPOF package (75% RDN through organic sources + two foliar spray of 10% cow urine and vermiwash at 30 and 50 DAS) which was comparable with rest of the treatments except control which showed the lowest dehydrogenase activity.

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